

Active Nematics for Mechanobiology: Patterning and Flows in Living Matter



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Abstract

Living matter is characterized by its ability to consume chemical energy to grow or exert active forces on its surroundings. Biological tissues composed of anisotropic and active cells can develop long-range orientational order with nematic symmetry. The constant injection of energy at the cellular level leads to intricate collective flows within the tissue. In this thesis, we apply the theory of active nematic hydrodynamics to explore the effects of cellular heterogeneity, chemical patterning, and mechanical confinement in living systems.

Using continuum simulations, we investigate a mixture of an active nematic fluid with a passive isotropic fluid. We demonstrate that the mixture spontaneously phase separates, forming micro-phases with distinct active and passive domains. This phase separation is dynamically driven by active forces at interfaces and can proceed even if it is energetically unfavourable. Introducing a second active species leads to complex alignment interactions between the two species. Mixtures of extensile and contractile nematics with elastic alignment show the strongest phase separation. Confining active nematic mixtures results in coherent spatial organization of the active component, with the resulting patterns depending on the orientation and flow fields inside the mixture, and the nematic orientation at the boundaries.

Flow patterns in an active nematic system are affected by external mechanical or chemical signals. Chemical patterns, in the form of Turing stripes, weaken and eventually dissolve in the presence of activity. Associating different stripes with extensile and contractile activities respectively can generate shearing flows inside each stripe, and long-time slipping of the stripes. On the other hand, confining active nematics with viscoelastic boundaries give rise to periodic flow reversals. This behaviour emerges from the interplay between the active forces driving fluid flow and the elastic restoring forces exerted by the confining walls.

Finally, we study breast cancer tumours inside an extracellular matrix as confined active matter. We show that active dynamics, rather than growth, dominate tumour behaviour. Our model accurately reproduces tumour morphologies obtained from clinical data and shows the effects of cancer clusters on fibre orientation in the extracellular matrix.

Overall, this thesis demonstrates how active matter physics can be a powerful tool to study biology. By exploring the interplay between active forces, mechanical properties, and environmental feedback, we highlight potential mechanisms for pattern formation and flows in living systems.

*To my grandmother, Dr. Tara Chatterjea, and my mother, Dr. Bhargavi
Chatterjea Bhattacharyya*

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“Would you tell me, please, which way I ought to go from here?”
 “That depends a good deal on where you want to get to.”
 “I don’t much care where.”
 “Then it doesn’t matter which way you go.”
 —Lewis Carroll (Alice in Wonderland)

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“The chess-board is the world; the pieces are the phenomena of the universe; the rules of the game are what we call the laws of Nature.”

— Thomas Henry Huxley

1

Introduction

When studying the mechanics of biological systems, it is necessary to account for the fact that cells are living. They generate ‘active’ forces, consuming nutrients to generate mechanical forces at a local level in the form of cell growth, motility, and apoptosis (cell death) [7]. Individual cells can move by changing their shape, and pushing and pulling on their surroundings [8, 9].

In many biological systems, however, cells are densely packed into tissues, forming confluent cell layers with no gaps between cells. These cells are connected to their neighbours using cell-cell junctions. Therefore, these cells can no longer move individually but are constrained to move collectively. This causes cells to jam, showing glass-like dynamics [10, 11]. However, many biological tissues undergo an unjamming transition, leading to cell layers behaving like a fluid [12, 13] over long timescales. Cells slide past each other by local cellular rearrangements (T1 transitions) [14], and the resultant collective cell motion is coherent over several cell lengths. Studying the collective behaviour of tissues can shed light on important open questions in biological mechanics [15].

One of the fundamental problems of mechanobiology is to study the growth of a developing embryo. During morphogenesis (‘morphe’ - form + ‘genesis’ - creation) [16], cells differentiate and migrate collectively to form complex structures such as the limbs, gut, heart and lungs of a developing organism. This process

involves careful coordination of different chemical signals and mechanical forces. Morphogenetic processes can be broadly categorized into two categories [16, 17]. The first category concerns cell proliferation and cell fate determination. These are mediated by chemical signalling, using proteins and genes as part of a complex biochemical framework. The second category involves processes which are more mechanical in nature. These include cell growth, collective cell migration, and cell sorting. Mechanical effects are relevant for cell packing [18], tissue folding in gastrulation [19], and budding in regenerative hydra [20]. Inter-cellular adhesion, cell motility and cellular interaction with the surrounding extracellular matrix all play an important role in governing the mechanics of morphogenesis [21, 22].

Another major challenge in mechanobiology is to understand how cancer cells migrate collectively [23]. Metastatic cancer cells can travel together collectively, maintaining cell-to-cell contacts. The specifics of this coordinated migration are influenced by both the specific cancer cell type [24], and also the properties of the local micro-environment [25]. Unravelling this complexity is crucial to understand the mechanisms underlying cancer metastasis.

Many theoretical models have been developed to study the mechanics of cell layers, at various levels of granularity and complexity [26]. A key feature of active models is that they are inherently out of equilibrium, with active forces constantly pumping energy into the system. One particularly successful model is the continuum theory of active nematics [27], which has recently generated considerable interest both as a model system out of thermodynamic equilibrium and as a theoretical model for cells.

Active nematic theory describes systems of anisotropic particles which apply local forces on their environment [27, 28] in a coarse-grained, continuum limit. It has been successfully used to model active turbulence in biological systems [29], spontaneous cellular flow under confinement [30], and the role of topological defects in growing bacterial biofilms [31] and regenerative hydra [32]. We will introduce the theory of active nematics in Chapter 2.

In this thesis, we have expanded on some of the implications of modelling cell layers as active nematics. This work aims to gain a deeper understanding of how cellular motility affects collective flows and patterning. We briefly touch on how active matter models can lead to new insights about cell sorting (Chapter 5) and metastatic cancer cells (Chapter 8).

The remainder of this thesis is structured as follows. In Chapter 2, we introduce the equations of active nemato-hydrodynamics. These equations describe the dynamics of the orientation field, and the flow field, and how changes in the orientation field and flow field are coupled to each other. Active nemato-hydrodynamics will serve as our model for biological tissue and provide the theoretical framework underpinning the results outlined in the rest of the thesis. The details about how we solve these equations numerically are delegated to Appendix A of this thesis.

In Chapter 3, we highlight the need for a multi-phase active model to study mixtures of different cell types. We then develop a two-fluid model with compressible components and active forcing, modifying a hybrid Lattice-Boltzmann algorithm to solve this system [33]. Using this approach, we show that homogeneous mixtures of active nematic and passive isotropic species spontaneously phase separate due to active forces, even if it is energetically unfavourable to do so. The phase separation emerges from the interplay of active nematic alignment and normal active forces at interfaces between different phases. Finally, we find the characteristic lengthscales of the resultant micro-phases and explain how the magnitudes and sizes of the phase-separated droplets depend on the thermodynamic free energy, viscous drag and surface tension of the two-fluid mixture.

In Chapter 4, we extend the results of the previous chapter to now study mixtures where both the species are active. Using numerical simulations and analytical arguments, we show that the orientation fields of two active species develop an effective alignment or anti-alignment interaction through the coupling due to viscous drag. We find that mixtures of species with opposite types of activity (extensile and contractile) show the highest phase separation with elastic alignment. We further study how the magnitude of phase separation and the morphology of

the phases can be controlled by changing the relative activities and concentration fractions of the mixture, or by increasing friction with the underlying substrate.

In Chapter 5, we look at mixtures of active nematics in confinement. While active nematics in bulk are always in a chaotic turbulent state [34], they can show a variety of flow states under confinement - these include spontaneous laminar flows inside channels [30], and spontaneous rotating flows in droplets [35]. We show that in confined mixtures of active and passive species, compressible active flows induce preferential aggregation of the active component in certain regions of the system. In a channel with laminar flows, the active component migrates towards the centre of the channel. In a droplet with spontaneous rotation, however, the active component sorts itself near the outer boundary. Even in active turbulent states under confinement, the active material preferentially accumulates at the boundary or the bulk depending on the details of thermodynamic or active anchoring at the walls. These results illustrate that active mechanisms of phase ordering may be relevant to cell sorting.

We then change gears in Chapter 6 to study the interplay of chemical signalling with active nematic cell layers. We first introduce Turing patterns [36], the archetype for chemical patterning, and couple them to active flows through advection by the nematic background. We find that active flows can affect the morphology and magnitude of Turing stripes, bending and breaking stripe segments. At high activities, the strength of the patterns decreases, ultimately dissolving past a critical activity. We show that the magnitude of patterning can be quantified by a single Karlovitz number, characterizing the relative strengths of active advection and the reaction flux. We further find that when the activity is dependent on the local concentration of morphogens, the system can self-organize into a pattern of stripes with internal shearing flows, with characteristic active instabilities making stripes fracture and slip sideways to join their neighbours. Our results show how mechanochemical coupling can lead to new dynamical states.

In Chapter 7, we look at mechanical feedback between active nematics and their confinement. We numerically study an active nematic channel confined by viscoelastic walls. The two regions are coupled through force balance and no-slip

boundary conditions at their common boundary. We find that this system exhibits periodic flow reversals over a range of activity and elasticity. This is an example of mechanical feedback from external confinement drastically altering the flow patterns of an active nematic in confinement.

In Chapter 8, we continue our investigation of confined active nematics, applying our model to study malignant breast cancer tumours inside a viscoelastic extracellular matrix (ECM). We find that our active model correctly reproduces the sizes and shapes of cancer tumours obtained from clinical data while making minimal assumptions. We further show that motility is the dominant feature driving the dynamics of these cancer tumours, and discuss how active cancer tumours create topological defects in the ECM and affect the orientation of ECM fibres close to tumour boundaries.

Throughout this thesis, we have used active nematics to show how patterning and flows are linked together in biology. In the process, we have discovered new active mechanisms for cell sorting, and seen how coupling active flows with chemical patterning and mechanical confinement can lead to new types of collective behaviour. We hope that this work has contributed to the broader goal of studying the physics of life, and shown that active matter theories can shed new light on the study of embryo morphogenesis and cancer metastasis.

“All models are wrong, but some are useful.”

— George E. P. Box

2

Active nematics

In this chapter, we introduce the theory of active nemato-hydrodynamics as a coarse-grained continuum description of confluent cell layers. These equations describe the orientation field, and the flow field, and how these fields couple to each other. Active nemato-hydrodynamics will serve as our model for biological tissue and provide the theoretical framework underpinning the results outlined in the rest of the thesis.

2.1 Introduction

Confluent cell layers are composed of motile cells which convert chemical energy into mechanical work, exerting active forces on their surroundings [37] (substrate and neighbouring cells). These cell layers can show a vast range of dynamical states, including glass-like jammed states [10, 11], coherent flocking states [38] and chaotic turbulent states [27, 39]. We now summarize different ways to mathematically model the dynamics of confluent cell layers.

A direct approach is to make a model that keeps track of the position, shape, internal configuration, and surroundings of each cell. Forces acting on cells from their surroundings can cause cells to move or deform. This is the idea behind the phase-field model [40, 41]. This model is computationally very intensive and scales poorly with the number of cells in the system.

A less computationally intensive method is to consider the confluent cells as polygons tessellating the cell layer. This sacrifices some detail about the cell packing and cell shape but simplifies the problem considerably to solving the dynamics of polygon vertices or polygon centres over time. Active forces between cells now act on the shared edges of polygons, or the polygon centres. This is the idea behind the vertex [42, 43] and Voronoi models [44, 45].

In order to study collective behaviour, one can coarse-grain even further describing the layer at lengthscales spanning several cell lengths. At this scale, the cell layer is described in terms of two fields: a velocity field $\mathbf{u}$ describing the collective motion of cells and a headless vector field $\mathbf{n}$ describing the average orientation of cells, defined at every region in space. The local active forces between cells are then modelled by active stresses inside the fluid. One such continuum description is the theory of active nematics [27, 34]. The advantage of a continuum model is that it is both computationally efficient and exactly solvable, leading to a deeper understanding of the physics underlying the dynamics of cell layers.

The rest of this chapter is structured as follows. In Sec. 2.2, we discuss the dynamics of the flow field $\mathbf{u}$ describing the collective flow of our model system. We follow this up by discussing the orientation field for a nematic liquid crystal in Sec. 2.3. Leveraging the head-tail symmetry of the constituent particles, we construct the nematic order parameter $\mathbf{Q}$, the Landau-de Gennes free energy for a liquid crystal, and discuss discontinuities in the nematic field, which are topological defects. Then, in Sec. 2.4, we couple the orientation field $\mathbf{Q}$ and flow field $\mathbf{u}$ to obtain the equation of nemato-hydrodynamics. In Sec. 2.5, we introduce activity in our model - showing how coarse-graining over many active particles producing dipolar forces gives rise to an active stress. We then discuss some of the consequences of having active stress - active instabilities in the bulk lead to a chaotic flow state called active turbulence, and active nematics at a boundary preferentially align at the interface (active anchoring). We conclude the chapter by briefly describing methods of numerically solving the model equations in Sec. 2.7, and finally summarizing the contents of this chapter in Sec. 2.8.

2.2 The Velocity Field

The velocity field $\mathbf{u}(\mathbf{r})$ describes the collective motion of the active nematic fluid at the location $\mathbf{r}$. We assume that the system is incompressible, and has a constant density. Conservation of mass then requires that the velocity field must be divergence-free

$$\nabla \cdot \mathbf{u} = 0. \quad (2.1)$$

Secondly, the fluid must satisfy the force balance condition

$$\rho(\partial_t + \mathbf{u} \cdot \nabla) \mathbf{u} = \nabla \cdot \mathbf{\Pi} - f\mathbf{u}. \quad (2.2)$$

The left-hand side of Eq.(2.2) is the convected derivative, which represents the total rate of change of momentum of a fluid element moving with the flow. The right-hand side denotes the sum of forces acting on the fluid, written in two parts - the derivative of a stress tensor $\mathbf{\Pi}$, consisting of all the forces which conserve the total momentum of the fluid, and a substrate friction term $f\mathbf{u}$, which describes momentum loss due to friction with the substrate.

The first term to add to the stress tensor is viscous dissipation. Fluid layers experience drag forces when they move past each other. These forces oppose the relative flow between layers and transfer momentum from the faster-moving layers to the slower layers. This can be modelled by a viscous stress

$$\mathbf{\Pi}^{visc} = 2\eta\mathbf{E}, \quad (2.3)$$

where η is the shear viscosity, and $\mathbf{E} = \frac{(\nabla\mathbf{u})+(\nabla\mathbf{u})^T}{2}$ is the strain rate tensor of the fluid.

Combining Eqs.(2.2) and (2.3) gives the Navier-Stokes equation, which describes a viscous Newtonian fluid. While tissues are viscoelastic [46], systems in which internal stress relaxation by T1 transitions [14], environmental remodelling [47] and growth [48] happen at a faster timescale than the timescales of collective tissue flows can be modelled as Newtonian fluids.

The ratio of inertial to viscous forces in a fluid is characterized by the Reynolds number, $Re \sim UL\rho/\eta$, where U and L are typical scales of velocity and length.

Biological systems are characterized by very small velocities U over microscopic lengthscales L , and so are always in a regime of vanishingly small Reynolds numbers. Hence, we can neglect the advective terms in favour of viscous forces in Eq.(2.2).

We will add many more terms in the stress tensor $\mathbf{\Pi}$ over the course of this thesis to model active forces, passive forces, and viscoelastic constitutive relations.

2.3 The Orientation Field

A collection of elongated particles with head-tail symmetry can form a phase of matter known as a nematic liquid crystal. Like liquids, nematics exhibit only short-range correlations between the positions of particles. Nematics also flow under the application of a shear stress. Unlike liquids, however, nematics prefer to align with their neighbours along their axis of elongation and show long-range orientational order, reminiscent of solid crystals [49]. At high concentrations, collections of elongated particles transform from an isotropic phase to a nematic liquid crystalline phase, due to entropic effects [50].

We coarse-grain the orientation fields of cell layers to model them as a nematic liquid crystal in a continuum description. This is done using the $\mathbf{Q}$ tensor formalism described below.

2.3.1 The $\mathbf{Q}$ tensor

The orientational field $\mathbf{n}(\mathbf{r})$, also known as the director field, describes the average axis of elongation of cells at the location $\mathbf{r}$. We further assume that cells have head-tail symmetry. This means that the system is unchanged under the operation $\mathbf{n} \rightarrow -\mathbf{n}$, i.e. the director field is headless. Unlike a normal vector, which is invariant under $\theta \rightarrow \theta + 2\pi$, the director field vector $\mathbf{n}$ is invariant under the operation $\theta \rightarrow \theta + \pi$ [49].

On coarse-graining, we also need to compute the degree of particle alignment with the director field $\mathbf{n}(\mathbf{r})$. For convenience, let us take the average orientation of the particles, $\mathbf{n}$, to lie along the x-axis. The orientation of each particle in a small volume around $\mathbf{n}$ can be written in terms of the polar angle θ in two dimensions. The

probability distribution of finding a particle with an orientation between $[\theta, \theta + d\theta]$ is therefore $f(\theta) d\theta$. Due to head-tail symmetry, f also satisfies $f(\theta) = f(\pi - \theta)$. The first non-zero term in the multipole expansion of $\theta(\mathbf{r})$ is given by [49]

$$S = \int_0^\pi f(\theta)(2 \cos^2 \theta - 1) d\theta. \quad (2.4)$$

If the particles are locally highly aligned, the distribution is sharply peaked around $\theta = 0$ ($f(\theta) = \delta(\theta)$), and the integral evaluates to $S = 1$. If the particles are randomly ordered, θ is uniformly distributed, and using some trigonometric identities, we find $S = 0$. Thus, S captures the magnitude of nematic order [49].

Finally, we can define the nematic tensor order parameter $\mathbf{Q}$, in d dimensions, as the second rank tensor given by [49]

$$\mathbf{Q} = \frac{d}{d-1} S \left(\mathbf{n} \mathbf{n} - \frac{\mathbb{I}}{d} \right). \quad (2.5)$$

This quantity contains all the information about the nematic director field. The Kronecker product $\mathbf{n} \mathbf{n}$ ensures head-tail symmetry, S captures the magnitude of nematic order, and the term proportional to the identity is subtracted to keep $\mathbf{Q}$ traceless. It will be convenient to write our equations in terms of $\mathbf{Q}$ instead of $\mathbf{n}$, since this explicitly preserves nematic symmetry.

In this thesis, we will only deal with two-dimensional systems ($d = 2$), in which case we can write down the components of the $\mathbf{Q}$ tensor explicitly as

$$\begin{aligned} Q_{xx} &= -Q_{yy} = S \cos(2\theta), \\ Q_{yx} &= Q_{xy} = S \sin(2\theta). \end{aligned} \quad (2.6)$$

However, many of the ideas presented in the thesis will carry over into three dimensions.

2.3.2 Landau de-Gennes Free Energy

In an ordered nematic state, the ground state of the system is modelled by a free energy functional with two terms - a bulk term to enforce local nematic ordering, and an elastic term to ensure long-range order in the orientational field. We write these conditions in terms of $\mathbf{Q}$ in the following subsections.

Bulk free energy

The scalar free energy in the order parameter $\mathbf{Q}$ can be expanded as [49]

$$f_{bulk} = \frac{\mathcal{A}}{2} \text{Tr}(\mathbf{Q}^2) + \frac{\mathcal{B}}{3} \text{Tr}(\mathbf{Q}^3) + \frac{\mathcal{C}}{4} \text{Tr}(\mathbf{Q}^2)^2 + \dots, \quad (2.7)$$

where $\mathcal{A}, \mathcal{B}$ and $\mathcal{C}$ are arbitrary coefficients which define the energy landscape in $\mathbf{Q}$ space. In two-dimensional systems, the term proportional to $\text{Tr}(\mathbf{Q}^3)$ vanishes [51] since the eigenvectors of Q are equal and opposite to each other. Hence, the bulk free energy simplifies to the form

$$f_{bulk} = C \left(S_{nem}^2 - \frac{\text{Tr}(\mathbf{Q}^2)}{2} \right)^2. \quad (2.8)$$

When $C > 0$ in Eq.(2.8), this describes a nematic with a magnitude of nematic order S_{nem} inside the bulk, which follows from minimizing the free energy. We will restrict ourselves to two dimensions, and use the form given by Eq.(2.8) for the rest of the thesis.

Typically at low concentrations of rod-like particles (nematogens) [52], an isotropic configuration with no nematic alignment is preferred primarily due to entropic reasons ($S_{nem} = 0$). However, at higher nematogen concentrations, the configuration which maximizes entropy has orientational alignment, but particles are free to move about in space ($S_{nem} \neq 0$) [50]. It is also possible to transition from the nematic configuration ($S_{nem} \neq 0$) to the isotropic configuration ($S_{nem} = 0$) by increasing the temperature past a critical temperature, called the nematic-isotropic transition temperature T_N .

Elastic free energy

In three dimensions, a nematic field can have three different types of distortions - bend, splay and twist, with elastic constants K_1, K_2 and K_3 respectively. This gives the Frank free energy [53]

$$f_{Frank} = \frac{K_1}{2} [\nabla \cdot \mathbf{n}]^2 + \frac{K_2}{2} [\mathbf{n} \cdot (\nabla \times \mathbf{n})]^2 + \frac{K_3}{2} [\mathbf{n} \times (\nabla \times \mathbf{n})]^2, \quad (2.9)$$

which penalizes distortions in the nematic field.

In this thesis, we will work under the assumption of a single elastic constant $K_1 = K_2 = K_3 \equiv K_0$, which has been used to accurately describe many biological systems [27, 31, 54]. The elastic free energy of a nematic can then be written as

$$f_{elastic} = \frac{K}{2} (\nabla \mathbf{Q})^2, \quad (2.10)$$

where $K = \frac{1}{8S_{nem}^2} K_0$. Moreover, twist deformations are impossible in two dimensions. The only allowed distortions of the orientation field are bends and splays (shown in Fig. 2.4).

Defects in the director field

The elastic free energy prefers smooth variations in the director field and typically prevents discontinuities. However, defects in the director field energy arise when domains with different orientations meet.

The winding number of defects is defined by integrating the director field angle along a closed contour around the defect,

$$m = \frac{1}{2\pi} \oint d\theta. \quad (2.11)$$

After going around a closed contour, the single-valuedness of the θ field requires that we return to the starting angle. By nematic symmetry, the director field is invariant after rotation by an integer multiple of π i.e. $\theta \rightarrow \theta + n\pi$, where n is any integer. Therefore, the winding number is allowed to take on any half-integer values $m = n/2$. This winding number is a topological quantity - and is the sum of the topological charges of the defects enclosed inside the integration contour. It can be non-zero only if the integration contour includes defects, and the total topological charge of a closed system is constant.

While in principle all defects with winding numbers m are allowed (where $2m$ can be any integer value), the elastic free energy for a defect of winding number m scales as $f_{defect} \propto Km^2$ [52]. Thus, defects with charges other than $\pm 1/2$ are energetically unfavourable and break apart into two or more $\pm 1/2$ defects, preserving charge conservation. Due to the conservation of topological charge, $\pm 1/2$ defects are created and annihilated in pairs.

The $+1/2$ defect is created when the director rotates counter-clockwise when travelling along the contour counter-clockwise. It has a characteristic comet shape, as shown in Fig. 2.1a. On the other hand, a $-1/2$ defect is observed when the director rotates clockwise when travelling along the contour counter-clockwise. It has a characteristic three-fold symmetry, as shown in Fig. 2.1b.

In three dimensions, grain boundaries form lines of discontinuity, known as disclination lines. Cross-sections of these disclination lines have complex defect structures which can be characterized into twist-like and wedge-like defects since the director field is allowed to rotate out of the plane. In this thesis, we will deal exclusively with comet-shaped $+1/2$ and trefoil-shaped $-1/2$ defects in two dimensions.

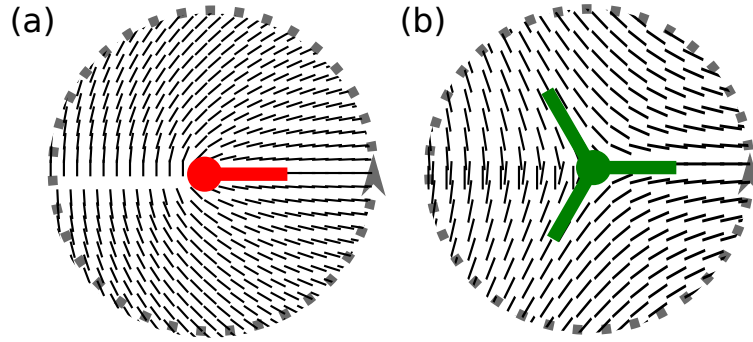


Figure 2.1: Nematic director fields show characteristic defects with winding numbers $+1/2$ and $-1/2$. Panel (a) shows the director field around a $+1/2$ defect, which has a comet-like structure with a head (marked by the red dot) and a tail (marked by the red line). In extensile active nematics, $+1/2$ defects self-propel towards the head, and the direction of motion is reversed in contractile active systems. Panel (b) shows the director field around a $-1/2$ defect, which has a three-fold symmetry (marked by the green lines). Active $-1/2$ defects are not self-propelled.

2.4 Nemato-hydrodynamics

In this section, we couple the velocity and orientation fields to find the dynamical equations describing a nematic liquid crystal.

The dynamics of the nematic field is given by the Beris-Edwards equation,

$$D_t \mathbf{Q} - \mathcal{W} = \Gamma \mathbf{H}. \quad (2.12)$$

The first term $D_t \mathbf{Q}$ is the advected derivative and describes the rate of change of $\mathbf{Q}$ which is moving with the flow. The second term $\mathcal{W}$ is the co-rotation term

which describes how the director field is rotated and stretched by vorticity and strain in the velocity field [27, 33, 55]. This is given by

$$\mathcal{W} = (\lambda \mathbf{E} + \mathbf{\Omega}) \cdot \left(\mathbf{Q} + \frac{\mathbb{I}}{d} \right) + \left(\mathbf{Q} + \frac{\mathbb{I}}{d} \right) \cdot (\lambda \mathbf{E} - \mathbf{\Omega}) - 2\lambda \left(\mathbf{Q} + \frac{\mathbb{I}}{d} \right) (\mathbf{Q} : \mathbf{E}), \quad (2.13)$$

where $\mathbf{E} = \frac{(\nabla \mathbf{u}) + (\nabla \mathbf{u})^T}{2}$ and $\mathbf{\Omega} = \frac{(\nabla \mathbf{u}) - (\nabla \mathbf{u})^T}{2}$ are the strain rate tensor and vorticity tensor of the fluid respectively, and λ is the flow-aligning parameter.

λ is a phenomenological parameter that describes the strength of alignment of the director axis with the strain rate in the fluid (see Fig. 2.2). For $\lambda = 0$, the orientation field only rotates with the vorticity but does not respond to the strain rate e.g. for isotropic circular particles. As the value of λ increases, the relative importance of the rate of strain tensor in Eq.(2.13) increases, corresponding to more elongated particles [55]. However, biological tissues have complicated micro-structure and may generate their own internal forces or align with flows in the environment to reduce strain [56, 57]. In this thesis, we have kept λ as a phenomenological parameter, which can be chosen based on the system of study.

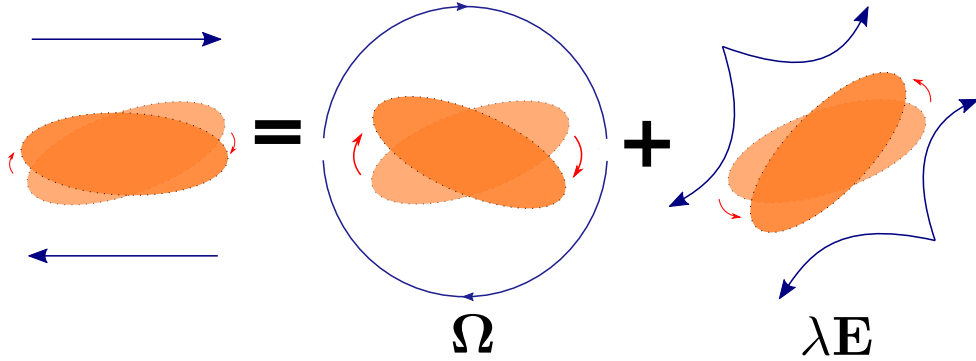


Figure 2.2: The response of the orientation field (orange) to the flow field (blue) has two components: rotation driven by the vorticity tensor $\mathbf{\Omega}$, and alignment with the stretching axis of the shear, described by the strain rate tensor $\mathbf{E}$. The relative importance of these terms is given by the flow-alignment parameter λ , which is a phenomenological parameter.

The final term in Eq.(2.12) describes the relaxation to the minimum of the free energy $\mathcal{F} = f_{bulk} + f_{elastic}$. This is governed by the molecular field,

$$\mathbf{H} = - \left(\frac{\delta \mathcal{F}}{\delta \mathbf{Q}} - \frac{\mathbb{I}}{d} \text{Tr} \left(\frac{\delta \mathcal{F}}{\delta \mathbf{Q}} \right) \right), \quad (2.14)$$

and Γ is the rotational diffusivity characterizing the relative importance of the free energy in the dynamics of $\mathbf{Q}$.

Distortions in the nematic field also generate passive stresses in the fluid field. This leads to restoring flows which tend to drive the nematic orientation back towards the free energy minimum. This is called the elastic backflow term and can be written as an additional term in the stress tensor in Eq.(2.2)

$$\begin{aligned} \Pi^{el} = & 2\lambda(\mathbf{Q} + \mathbb{I}/d)(\mathbf{Q} : \mathbf{H}) - \lambda \mathbf{H} \cdot (\mathbf{Q} + \mathbb{I}/d) - \\ & \lambda(\mathbf{Q} + \mathbb{I}/d) \cdot \mathbf{H} - (\nabla \mathbf{Q}) \cdot \frac{\partial \mathcal{F}}{\partial \nabla \mathbf{Q}} + \mathbf{Q} \cdot \mathbf{H} - \mathbf{H} \cdot \mathbf{Q}. \end{aligned} \quad (2.15)$$

2.5 Adding Activity

So far, we have described a nematic liquid crystal with coupled velocity and director fields. In this section, we account for the forces exerted by nematics on their surroundings, modelled by active stress. We then look at some consequences of having such an active term.

Force Dipoles

Since nematics have head-tail symmetry, the locally generated forces by an active nematic must also have head-tail symmetry. Moreover, even when cells produce unidirectional polar forces, the confluent layer behaves like an active nematic if cell-cell interactions are dominant [58], or under fluctuating polar forces [59].

The simplest model for this nematic active forcing is a point force dipole, with equal and opposite forces acting on each end of the elongated nematic axis. We shall coarse-grain these local active forces into an active stress term. Let us consider a system composed of many active nematic particles, each of length $2l$. An active nematic particle labelled by the index i has its centre located at $r^{(i)}$, and an orientation given by $\mathbf{a}^{(i)}$ (see Fig. 2.3). Each particle generates equal

and opposite forces at each end with constant magnitude F_ζ . The net active force at position $\mathbf{r}$ is given by [34]

$$\nabla \cdot \mathbf{\Pi}^{act}(\mathbf{r}) = \mathbf{F}^{act}(\mathbf{r}) = F_\zeta \sum_i \mathbf{a}^{(i)} \left[-\delta(\mathbf{r} - \mathbf{r}^{(i)} + l\mathbf{a}^{(i)}) + \delta(\mathbf{r} - \mathbf{r}^{(i)} - l\mathbf{a}^{(i)}) \right], \quad (2.16)$$

where $\mathbf{\Pi}^{act}$ is the active stress, $\mathbf{F}^{act}$ is the active force, and the index i sums over the particles being coarse-grained, with locations $\mathbf{r}^{(i)}$ close to $\mathbf{r}$. Since the particle size is much smaller than the coarse-graining scale, $l \ll \mathbf{r} - \mathbf{r}^{(i)}$, we can approximate Eq.(2.16) as

$$\nabla \cdot \mathbf{\Pi}^{act}(\mathbf{r}) = \mathbf{F}^{act}(\mathbf{r}) \approx -F_\zeta (2l) \nabla \left[\sum_i \mathbf{a}^{(i)} \mathbf{a}^{(i)} \delta(\mathbf{r} - \mathbf{r}^{(i)}) + \dots \right]. \quad (2.17)$$

We recognize the tensor order parameter $\mathbf{Q}$ in Eq.(2.17). After summing over all the force dipoles in an unit volume, the nematic component of the active stress tensor coarse-grains to

$$\mathbf{\Pi}^{act}(\mathbf{r}) = -\phi(\mathbf{r}) F_\zeta (2l) \left(\mathbf{n} \mathbf{n} - \frac{\mathbb{I}}{d} \right) = -\zeta \phi(\mathbf{r}) \mathbf{Q}(\mathbf{r}). \quad (2.18)$$

Here $\phi(\mathbf{r})$ denotes the local number density of active nematic particles, $\mathbf{Q}(\mathbf{r})$ is the nematic order parameter, and we have now introduced an activity parameter ζ which characterizes the magnitude of each local force dipole. If the concentration is constant everywhere in space, we may absorb it into the activity parameter ζ to find the active stress [27]

$$\mathbf{\Pi}^{act} = -\zeta \mathbf{Q}. \quad (2.19)$$

Since the nematic tensor $\mathbf{Q}$ contains all information about the orientation field, any nematic active stress can be expressed in terms of $\mathbf{Q}$ and its gradients. The simplest term, proportional to $\mathbf{Q}$ itself, corresponds to the coarse-graining of active force dipoles, as we have shown above.

Finally, we express the total stress tensor for an active nematic fluid

$$\mathbf{\Pi} = \mathbf{\Pi}^{visc} + \mathbf{\Pi}^{el} + \mathbf{\Pi}^{act} \quad (2.20)$$

where $\mathbf{\Pi}^{visc}$ (Eq. 2.3), $\mathbf{\Pi}^{el}$ (Eq. 2.15), and $\mathbf{\Pi}^{act}$ (Eq. 2.19) are the viscous, elastic and active stresses respectively.

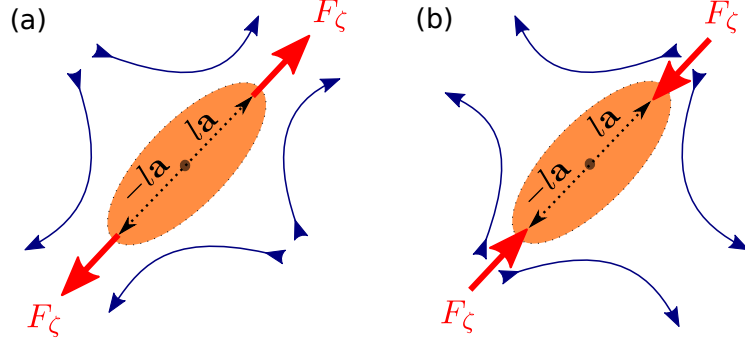


Figure 2.3: Cells exert active forces on their environment. For active nematic force dipoles, these can be categorized as **(a)** extensile (pusher, $\zeta > 0$) or **(b)** contractile (puller, $\zeta < 0$) forces (shown in red). The blue arrows show the flows driven by these active forces.

The sign of ζ in Eq.(2.19) picks the direction of active forces in the model. If $\zeta > 0$, the active forces point outwards along the long axes, creating pushing forces. These are known as extensile particles. On the other hand, if $\zeta < 0$, the active forces create pulling forces along the elongation axis. These are contractile particles. The forces and flows associated with extensile and contractile particles are shown in Fig. 2.3 .

Next, we look at some of the consequences of active forces in nematic liquid crystals.

2.5.1 Active turbulence

The equations of active nematic hydrodynamics couple the orientation field and flow field. Gradients in the orientation field give rise to active forces, setting up gradients in the flow field. Gradients in the flow field, in turn, rotate the director field changing the active forces in the system. In this subsection, we look at how active dipolar forces in bulk can lead to a chaotic flow state known as active turbulence.

Hydrodynamic instabilities

The lowest energy configuration of a nematic liquid crystal is a perfectly ordered state, with the director field aligned parallel everywhere and with no topological defects. In the absence of activity, this state is stable under perturbations - any small distortion in the nematic field creates elastic restoring forces which drive the system back to the aligned phase.

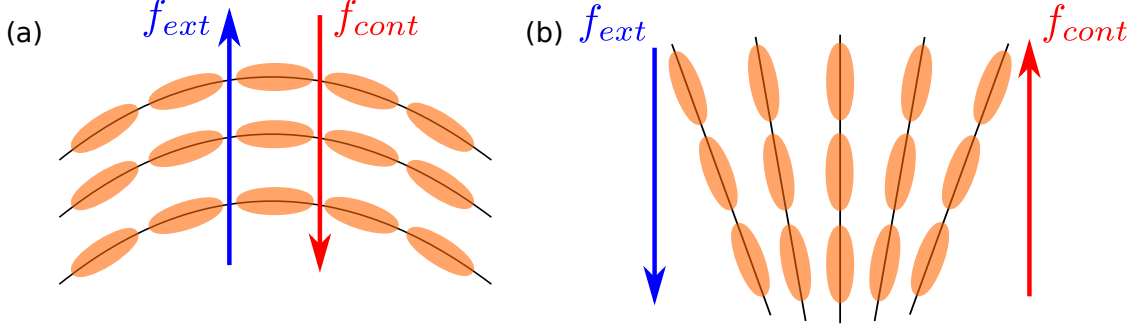


Figure 2.4: The typical distortions in two-dimensional nematics are bend (shown in panel (a)) and splay (shown in panel (b)). The blue arrow shows the direction of active forces if the nematics are extensile, while the red shows the direction of contractile active forces.

However, on adding activity, certain perturbations from the ordered state become unstable. For an extensile nematic, bend perturbations in the director field lead to an imbalance of active forces in the fluid, setting up flows which then increase the perturbation even further [34]. For contractile nematics, the same is true for splay perturbations instead (Fig. 2.4).

In both cases, these instabilities lead to regions of bend or splay growing sharper, forming walls with sharp distortions in the director field. These walls eventually lead to the nucleation of new $\pm 1/2$ defects. Since $+1/2$ defects are motile, active forces then push these defects away from each other, leading to the spontaneous creation of topological defects and active flows inside the bulk, pushing the system further away from the aligned ground state and into active turbulence [60].

Flows around defects

As we have discussed, nematic liquid crystals can have topological defects with charge $\pm 1/2$. In a passive nematic, elastic forces cause these defects to be attracted to each other, and annihilate in pairs. In active systems, however, the distortions in the director field lead to characteristic active flows around each type of defect.

For a $+1/2$ defect, the comet shape of the director field breaks directional symmetry. This generates unidirectional flows which self-propel the defect [61]. For extensile nematics, $+1/2$ defects move in the tail-to-head direction. For contractile nematics, $+1/2$ defects self-propel in the head-to-tail direction.

For a $-1/2$ defect, three-fold symmetry ensures that the defect cannot self-propagate in any given direction [61]. However, the distortions in the director field still generate flows around the defect in a flower-like pattern. The direction of the flows around a $-1/2$ defect for extensile and contractile nematics is opposite since the active forces point in the opposite direction.

Active Turbulence

As we have seen, the uniformly aligned nematic state is unstable to activity. As a result, active nematics have out-of-equilibrium steady states with chaotic patterns in the director and flow fields. This state is characterized by a finite number of topological defects, which are constantly created and annihilated at different locations. This dynamical state is known as active turbulence (see Fig. 2.5) [27, 34]. Although the dynamics may seem chaotic, the state we shall refer to as “active turbulence” has underlying structure in the correlation functions of the director and velocity field [39].

In active turbulence, the elastic and active stresses in the fluid balance. The magnitude of elastic stresses scales inversely with the lengthscale of distortions $\Pi^{el} \propto Kl^{-2}$, while the active stress is independent of lengthscale $\Pi^{act} \propto |\zeta|$. Active turbulence forms vortices at the characteristic lengthscale where these balance, given by the active lengthscale $l_{act} = \sqrt{K/|\zeta|}$. The typical lengthscale of the bend and splay instabilities is also the active lengthscale [39, 62, 63].

Active turbulence also has a characteristic correlation timescale $t_{act} = \eta/\zeta$, which decreases with increasing the magnitude of activity. This notion of characteristic lengthscales and timescales is different from the picture of turbulence in inertial systems, with energy transfer between scales. Active turbulence has both energy injection and dissipation at the active lengthscale, and there is no energy transfer between lengthscales unlike inertial turbulence [39].

While active nematics in bulk are always turbulent, different flow states are observed in confined active nematics, when the lengthscales of confinement are

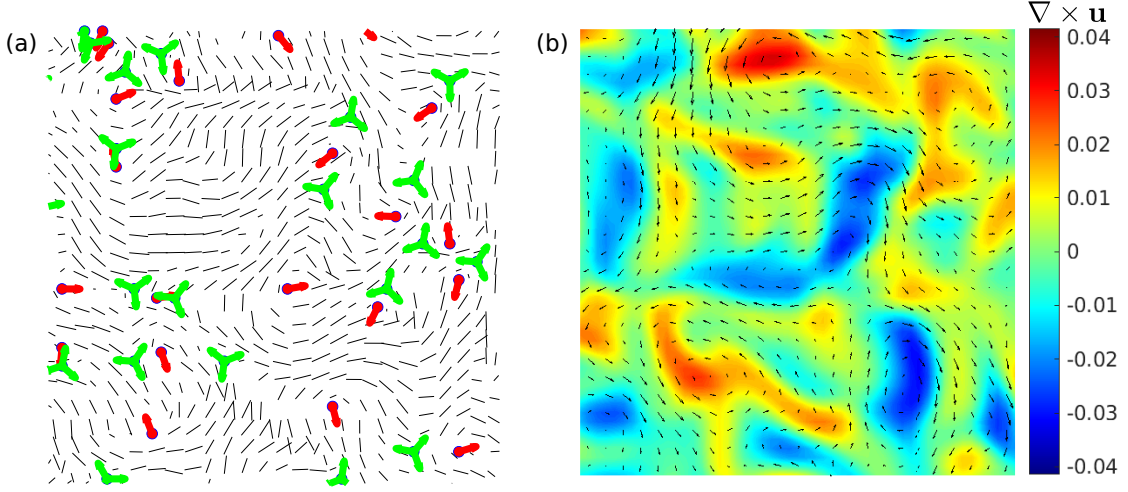


Figure 2.5: A typical snapshot of an active nematic undergoing active turbulence. Panel (a) shows the chaotic nature of the orientation field, with $+1/2$ and $-1/2$ defects labelled in red and green respectively. Panel (b) shows the underlying velocity and vorticity field of this snapshot. The black arrows show the direction of the velocity, while the colourbar shows the magnitude of the vorticity.

comparable to this emergent active lengthscale. We will explore confined active nematics further in Chapters 5, 7 and 8.

2.5.2 Active anchoring

Another consequence of nematic active forces is that the orientation field aligns parallel or perpendicular to interfaces and boundaries. This effect is called ‘active anchoring’.

Active anchoring arises from an imbalance of active forces at the interface between active and passive fluids. Let us consider that the active nematic director field has an angle θ with respect to an interface. The direction of the nematic is $\mathbf{n}$, and the normal and tangent to the interface are $\mathbf{m}$ and $\mathbf{l}$ respectively (see Fig. 2.6a). There is a difference in the nematic concentration ϕ (defined in Eq.(2.18)) and nematic order S across the interface. This leads to active forces at the interface [64] given by

$$\mathbf{F} = -\zeta|\nabla(S\phi)|[2(\mathbf{m} \cdot \mathbf{n})\mathbf{n} - \mathbf{m}], \quad (2.21)$$

with tangential component $F^{tang} = 2\zeta|\nabla(S\phi)|(\mathbf{m} \cdot \mathbf{n})(\mathbf{l} \cdot \mathbf{n})$ and normal component $F^{norm} = -\zeta|\nabla(S\phi)|(2(\mathbf{m} \cdot \mathbf{n})^2 - 1)\mathbf{m}$. The tangential component of the force at the interface creates flows which rotate the director field [64] (see Fig. 2.6b).

The tangential forces are zero if the director field is parallel or perpendicular to the interface, and change direction when the directors rotate past the parallel or perpendicular configuration.

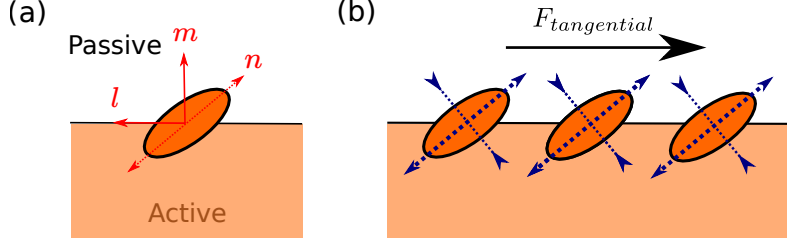


Figure 2.6: (a) Schematic showing an active nematic at a boundary, with relevant directions marked. (b) Extensile ($\zeta > 0$) active nematics at an interface generate a tangential force which aligns them parallel to the boundary. The forces for contractile ($\zeta < 0$) nematics point in the opposite direction, tending to align nematics perpendicular to the boundary.

If the activity is extensile, the active flows rotate the director to be parallel to the interface. This is known as planar anchoring. For contractile activity, however, the active flows rotate the director in the opposite direction, preferring the orientation perpendicular to the interface. This is called homeotropic anchoring. Active anchoring at boundaries and interfaces plays an important role in controlling orientation and flows in active nematics, and we will return to this idea many times over the course of this thesis.

2.6 Phase-Field Modelling

In a system with distinct phases of different properties, a single phase-field φ can be used to describe the different regions of the simulation domain. We will define $\varphi = 1$ to be inside an active region, and $\varphi = 0$ to be outside the active region. The mixing free energy is $\mathcal{F}_\varphi = \int f_\varphi dA$, where dA is an area element and the free energy density of the phase field f_φ is given by

$$f_\varphi = \frac{1}{2}A_\varphi \varphi^2(1 - \varphi)^2 + \frac{1}{2}\kappa_\varphi (\nabla\varphi)^2. \quad (2.22)$$

The first term in the free energy ensures that the system self-organizes into regions with $\varphi = 0$ and $\varphi = 1$. The second term penalizes sharp changes in φ . At the

boundaries between regions, φ transitions smoothly from 0 to 1 over an interface lengthscale $l_{int} \approx \sqrt{\frac{\kappa_\varphi}{A_\varphi}}$ [65].

The dynamics of φ is given by the Cahn-Hilliard equation [66]

$$\frac{\partial \varphi}{\partial t} + (\mathbf{u} \cdot \nabla) \varphi = \Gamma_\varphi \nabla^2 \tilde{\mu}, \quad (2.23)$$

where the chemical potential is $\tilde{\mu} = \frac{\delta \mathcal{F}_\varphi}{\delta \varphi}$, and Γ_φ is the mobility coefficient. The left-hand side describes how φ changes while being advected by the flow, while the right-hand side describes how φ responds to gradients in the free energy. The interface between the two phases has a surface tension given by $\gamma = (1/6)\sqrt{A_\varphi \kappa_\varphi}$ [67].

2.7 Simulation Methods

We simulate an active nematic system numerically by solving the flow field $\mathbf{u}$ (Eqs.(2.1) and (2.2)) and the orientation field $\mathbf{Q}$ (Eq.(2.12)) simultaneously. In the literature, the $\mathbf{Q}$ field is usually solved using either a finite-difference (FD) or a finite-element method (FEM) [68]. There are many different methods to solve for the flow field - pseudo-spectral methods [69] and the lattice-Boltzmann method [52] are the most commonly used.

We simulate the system using a hybrid Lattice-Boltzmann (LB) method [33] on a fixed Cartesian grid. This uses a Lattice-Boltzmann method to solve for the flow field, Eq.(2.2), with a BGK collision operator and additional source terms to model forces and stresses [52]. For convenience of numerical simulations, the simulated system is isothermal instead of exactly incompressible, and we include the advective term in Eq.(2.2). However, numerical fluctuations of fluid density are resolved by sound waves which are two orders of magnitude faster than the other timescales in the system [67]. Thus, the simulated system is effectively incompressible (with an error $\sim 0.01\%$). Moreover, while we include the inertial terms in the simulation, the Reynolds number of the system is $Re \approx 0.003$ - ensuring that the viscous terms dominate the inertial terms in Eq.(2.2).

Simultaneously to the fluid field, we solve the dynamics of the nematic tensor $\mathbf{Q}$ using a finite difference method, as described in Refs. [33, 70]. We used a

five-point finite-difference stencil to approximate derivatives numerically [71], and verified accuracy with multiple finite-difference steps and a predictor-corrector method [68]. We use simulation units where the lengthscales and timescales are normalized to 1, these are referred to as LB units. Further details of the hybrid Lattice-Boltzmann method are described in Appendix A.

2.8 Conclusions

In this chapter, we have described how active nematics naturally arise as a coarse-grained description of motile cell layers. We modelled this system with a collective nematic tensor field $\mathbf{Q}$, and a collective flow field $\mathbf{u}$. We discussed the dynamics of these components individually and showed how they are coupled to each other through the theory of nemato-hydrodynamics.

Finally, we introduced activity to model forces exerted by the cells on their surroundings. We coarse-grained these forces to derive the active stress, and discussed self-propelled defects, active turbulence and active anchoring as natural consequences of this theory.

Throughout the rest of this thesis, we use active nematics as the model for living matter.

Take-home message:

Active nematics are a theoretical model to study living cells.

“Rājāra hasta karē samasta kāñālēra dhana curi. ”
(Translation: *The king’s hand steals all the poor’s*
wealth.)

— Rabindranath Tagore (Dui Bigha Jomi)

3

Phase Separation driven by Active Flows

In this chapter, we highlight the need for a multi-phase active model to study mixtures of different cell types. We then develop a two-fluid model with compressible components and active forcing, and modify the hybrid Lattice-Boltzmann algorithm to simulate this system. Using this model, we show that homogeneous mixtures of active nematic and passive isotropic species spontaneously phase separate due to active forces. The mechanism relies on the interplay of active anchoring and normal active forces at interfaces.

3.1 Introduction

Phase separation is a ubiquitous phenomenon found across a wide variety of biological systems. Inside cells, membrane-less organelles such as stress granules and nucleoli phase separate from their surroundings to allow different chemical environments for biochemical reactions [72, 73]. Different cell types sort themselves into distinct regions in confluent layers [74, 75], and during growth and morphogenesis [16, 76] (see Fig. 3.1). Bacterial colonies also undergo segregation, with species of different phenotypes clustering together [77]. We will use the terms ‘phase separation’, ‘segregation’, ‘demixing’, and ‘phase ordering’ interchangeably to refer to this phenomenon, as has been used in the literature.

A large body of research has looked at biological phase separation through the lens of equilibrium thermodynamics, attributing the ordering to the minimization of a free energy. A notable thermodynamic model for cell sorting is the differential adhesion hypothesis (DAH) [78, 79], which proposes that cells preferentially adhere to other cells of the same type because of differences in surface tension between like and unlike cells. Other thermodynamic approaches include considerations of line tension [74, 80] and surface contraction [74, 81] of cells. However, biological matter is inherently out of thermodynamic equilibrium - which opens the possibility of phase separation mechanisms that are outside the realm of free energy minimization principles [82].

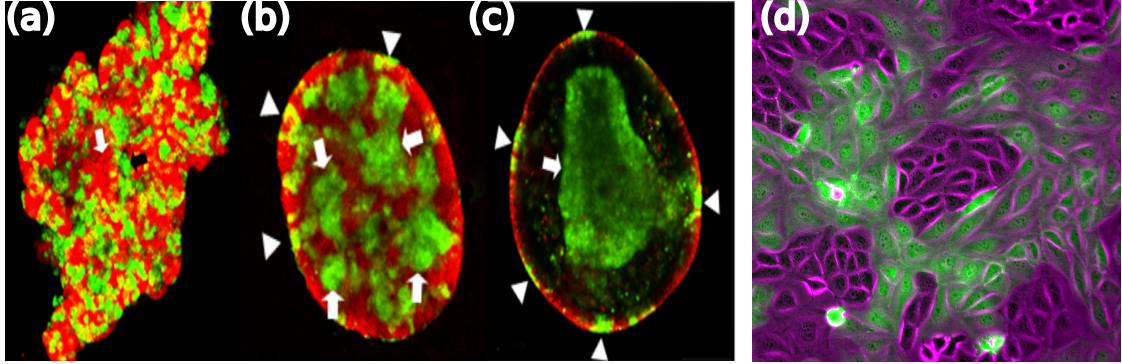


Figure 3.1: Cell Sorting: (a), (b), (c) starfish embryo at times (a) $t = 4$ hr, (b) $t = 8$ hr, (c) $t = 48$ hr showing sorting of embryonic cells (adapted from Ref [75]). (d) Phase ordering of extensile (magenta) and contractile (green) epithelial MDCK cells (courtesy of L. Balasubramaniam [83]).

Active systems can show examples of non-equilibrium phase separation. Motility-induced phase separation (MIPS) [84, 85] is an example of active phase separation, where self-propelled particles can become trapped in regions of high density thus forming a dense phase and a dilute phase. Many other novel mechanisms of ordering in active systems have been reported in the literature, including aligning torques [86], bond formation and breaking [87], control of boundary conditions [88], and the hydrodynamic interactions between dumbbell-shaped swimmers [89].

It is still not clear whether and how active stresses can lead to phase ordering in a binary mixture of an active nematic and a passive fluid in the absence of any thermodynamic driving force. To answer this question, we extend the continuum

theories of active nematohydrodynamics to a two-fluid model with separate velocity fields, coupled through viscous drag, for each fluid component [90, 91]. This allows us to model relative motion between the two fluids, which is a requirement for activity-driven phase ordering. Two-fluid models that have been used in biological contexts include studies of cytoskeleton dynamics and growing biofilms [92–95].

The rest of this chapter is organized as follows. In Sec. 3.2, we derive the constitutive equations for a two-fluid active nematic model with compressible components. We then simulate this model in Sec. 3.3, showing that homogeneous mixtures of active nematic and passive isotropic species spontaneously phase separate. In Sec. 3.3.1, we investigate the mechanism in detail, showing that the phase separation emerges from the interplay of active anchoring and normal active forces at interfaces. We explore how the magnitude of phase separation and the domain sizes depend on the activity, free energy, surface tension, and viscous drag between fluids in Sec. 3.4. The system has a minimum critical activity required for phase separation, as shown in Sec. 3.5. We conclude this chapter by studying how domain curvature affects the mechanism of phase separation in Sec. 3.6.

3.2 Model

We study a mixture of two active fluids [91, 93, 96]. In this chapter, we will work in index notation for convenience. Each fluid component has a local density ρ^i , velocity field u_α^i , and chemical potential μ^i , where Latin superscripts $i = 1, 2$ index the different fluids, and the Greek subscripts $\alpha = 1, 2$ denote the spatial directions. Summation convention is used for the Greek indices but will be specified explicitly for the Latin indices when applicable.

Each component fluid obeys the mass continuity equation

$$\partial_t \rho^i + \partial_\alpha \rho^i u_\alpha^i = 0 \quad (3.1)$$

and the momentum balance equation (Eq.(2.2))

$$\begin{aligned} \partial_t \rho^i u_\alpha^i + \partial_\beta \rho^i u_\alpha^i u_\beta^i = & -\rho^i \partial_\alpha \mu^i + F_\alpha^{visc,i} + F_\alpha^{body,i} \\ & -f \phi^i u_\alpha^i + \gamma \phi(1 - \phi)(u_\alpha^{3-i} - u_\alpha^i), \end{aligned} \quad (3.2)$$

where

$$\begin{aligned}\rho^c &= \rho^1 + \rho^2, \\ \phi^1 &= \phi = \rho^1 / \rho^c, \\ \phi^2 &= 1 - \phi = \rho^2 / \rho^c\end{aligned}\tag{3.3}$$

are the total density, concentration fraction of component 1, and concentration fraction of component 2 respectively. In Eq. (3.2), the left-hand side denotes the convective derivative of the fluid momentum density ($\rho^i u_\alpha^i$), while the right-hand side describes the force acting on the fluid per unit volume. The forces are modelled by a thermodynamic force ($-\rho^i \partial_\alpha \mu^i$), a viscous drag between the component fluids $\gamma \phi(1 - \phi)(u_\alpha^{3-i} - u_\alpha^i)$, an internal viscous dissipation for each fluid $F_\alpha^{visc,i}$, a substrate friction term $-f \phi^i u_\alpha^i$, and a body force $F_\alpha^{body,i}$ which models the local forces generated by nematic stresses.

We formulate our equations to treat this system as an incompressible fluid with compressible components. In order to do so, we define new velocity fields

$$\begin{aligned}u_\alpha^c &= \phi u_\alpha^1 + (1 - \phi) u_\alpha^2, \\ \delta u_\alpha &= u_\alpha^1 - u_\alpha^2,\end{aligned}\tag{3.4}$$

which are the centre of mass velocity of the total fluid and relative flow between the fluids respectively. We reserve the superscript c to refer to the combined (centre of mass) fluid. Moving forward, we will assume that the relative flow is much smaller than the combined velocity of the fluid i.e. $|\delta \mathbf{u}| \ll |\mathbf{u}^c|$. We verify a posteriori that this assumption holds true in our simulations, where we find $|\delta \mathbf{u}|/|\mathbf{u}^c| \leq 0.1$, with typical values of the ratio around 0.05 .

In order to proceed further, we will transform to the new coordinates by repeatedly making use of the following identities:

$$\begin{aligned}\rho^1 u_\alpha^1 + \rho^2 u_\alpha^2 &= \rho^c u_\alpha^c, \\ u_\alpha^c - \phi \delta u_\alpha &= u_\alpha^2, \\ u_\alpha^c + (1 - \phi) \delta u_\alpha &= u_\alpha^1.\end{aligned}\tag{3.5}$$

Adding Eqs. (3.1), (3.2) for each component gives the equations of motion for the combined fluid [97, 98]:

$$\partial_t \rho^c + \partial_\alpha \rho^c u_\alpha^c = 0, \quad (3.6)$$

$$\begin{aligned} \partial_t \rho^c u_\alpha^c + \partial_\beta \rho^c u_\alpha^c u_\beta^c = \sum_{i=1}^2 \Big[-\rho^i \partial_\alpha \mu^i + \\ F_\alpha^{visc,i} - f \phi^i u_\alpha^i + F_\alpha^{body,i} \Big]. \end{aligned} \quad (3.7)$$

Here, we have neglected the term $\rho^c \phi (1 - \phi) \delta u_\alpha \delta u_\beta$ on the left-hand side of Eq.(3.7), since it is of order $(\delta \mathbf{u})^2$. Notice that the viscous drag between the components drops out of the equation for the combined fluid. The combined fluid conserves mass (Eq.(3.6)) and momentum (Eq.(3.7)) and acts as a typical incompressible fluid. We simulate this fluid using the Lattice-Boltzmann method described in Sec. 2.7.

We now discuss the terms on the right-hand side of the momentum balance equation (3.7) in turn. The first term is the thermodynamic force, which follows from a Ginzburg-Landau free energy functional of the form [97, 98]

$$\mathcal{F}_{LG} = \int d^2 \mathbf{r} \left(\psi(\rho^1, \rho^2) + \frac{1}{2} \kappa ||\nabla \phi||^2 \right) \quad (3.8)$$

where

$$\psi = \frac{1}{3} \rho^c \ln \rho^c + \rho^c \left\{ a \left(\phi - \frac{1}{2} \right)^2 + b \left(\phi - \frac{1}{2} \right)^4 \right\}. \quad (3.9)$$

The first term of (3.9) corresponds to an increase in the pressure when the combined fluid density increases, resulting in fluid flows (sound waves) that equalize the local density much faster than the timescales of active flows. This results in the system being practically incompressible ($\leq 0.01\%$). The second term is a Landau free energy which drives the system to a uniformly mixed configuration if $a \geq 0, b \geq 0$. The chemical potentials for each fluid are defined as $\mu^i = \delta \mathcal{F}_{LG} / \delta \rho^i$. We can write [97, 98] the chemical potential term for the combined fluid $\sum_{i=1}^2 -\rho^i \partial_\alpha \mu^i$ as the divergence of a stress tensor $-\partial_\beta \sigma_{\alpha\beta}^{thermo}$, where

$$\sigma_{\alpha\beta}^{thermo} = p \delta_{\alpha\beta} + \kappa (\partial_\alpha \partial_\beta \phi - \frac{1}{2} ||\nabla \phi||^2 \delta_{\alpha\beta}), \quad (3.10)$$

$$-p = \psi - \frac{\partial \psi}{\partial \rho^1} \rho^1 - \frac{\partial \psi}{\partial \rho^2} \rho^2. \quad (3.11)$$

$p = \rho^c / 3$ is an isotropic pressure consistent with an isothermal equation of state. The term in κ is an anisotropic stress resulting from the surface tension between the two component fluids.

The second term on the right-hand side of Eq. (3.7) is the usual viscous stress (Eq.(2.3)), defined by

$$F_{\alpha}^{visc,i} = \partial_{\beta} \rho^i \nu^i (\partial_{\alpha} u_{\beta}^i + \partial_{\beta} u_{\alpha}^i - \delta_{\alpha\beta} \partial_{\gamma} u_{\gamma}^i). \quad (3.12)$$

Here, we have written the viscosity η^i in terms of the kinematic viscosity ν^i . In the following, we will choose $\nu^1 = \nu^2$ in order to avoid viscosity gradients and associated instabilities [99] in the combined fluid.

The third term on the right-hand side of Eq. (3.7) describes the friction between each component fluid and the substrate.

Finally, the last term in Eq. (3.7) is the body force acting locally on the fluid at each point which arises from passive and active nematic stresses $F_{\alpha}^{body,i} \equiv F_{\alpha}^{Q,i}$. We next discuss the dynamics of each active nematic species and the form of $F_{\alpha}^{Q,i}$.

Each fluid component i is a nematic liquid crystal [26, 27, 34] described by a symmetric traceless tensor [49] (Eq.(2.5))

$$Q_{\alpha\beta}^i = S_{nem}^i (2n_{\alpha}^i n_{\beta}^i - \delta_{\alpha\beta}) \quad (3.13)$$

in 2D where n_{α}^i is a headless vector denoting the orientation of the local nematic order, called the director field, and S_{nem}^i is the magnitude of the nematic order. The order parameter relaxes towards the minimum of the Landau-de Gennes free energy density [27, 49] (Eqs.(2.10), (2.8))

$$\mathcal{F}^{LdG,i} = C^i ((S_{nem}^i)^2 - \frac{1}{2} Q_{\alpha\beta}^i Q_{\alpha\beta}^i)^2 + \frac{K^i}{2} \partial_{\gamma} Q_{\alpha\beta}^i \partial_{\gamma} Q_{\alpha\beta}^i \quad (3.14)$$

defined so that the minimum free energy corresponds to a state with an order parameter of magnitude S_{nem}^i , with the director uniformly aligned in space.

We couple the orientation fields of the two species explicitly by adding an extra aligning or anti-aligning interaction in the free energy of the form

$$\mathcal{F}^{Q_1,Q_2} = -L_{12} \phi(1 - \phi) Q_{\alpha\beta}^1 Q_{\alpha\beta}^2. \quad (3.15)$$

When both species are mixed together, this term aligns the director fields for $L_{12} > 0$, and anti-aligns them for $L_{12} < 0$. When both species are well aligned the

effect of this term is just to renormalize the coefficient a in Eq.(3.9). Thus, we have not considered this term separately in the chemical potential. From Eqs. (3.8), (3.14) and (3.15) the total free energy of the system is given by

$$\mathcal{F} = \mathcal{F}_{LG} + \mathcal{F}^{LdG,1} + \mathcal{F}^{LdG,2} + \mathcal{F}^{Q_1,Q_2}. \quad (3.16)$$

We now describe the dynamics of the orientation field. We expect the director field to not only be advected by the fluid but also to be rotated according to the vorticity tensor $\Omega_{\alpha\beta}^i = (\partial_\alpha u_\beta^i - \partial_\beta u_\alpha^i)/2$. Additionally, the director may tend to orient along the strain rate tensor $E_{\alpha\beta}^i = (\partial_\alpha u_\beta^i + \partial_\beta u_\alpha^i)/2$ of the fluid. The response to gradients of the flow is modelled by the co-rotation term [27, 49] (Eq.(2.13))

$$\begin{aligned} S_{\alpha\beta}^i = & (\lambda^i \tilde{E}_{\alpha\chi}^i + \Omega_{\alpha\chi}^i)(Q_{\chi\beta}^i + \delta_{\chi\beta}/2) + \\ & (Q_{\alpha\chi}^i + \delta_{\alpha\chi}/2)(\lambda^i \tilde{E}_{\chi\beta}^i - \Omega_{\chi\beta}^i) \\ & - 2\lambda^i(Q_{\alpha\beta}^i + \delta_{\alpha\beta}/2)Q_{\chi\gamma}^i \tilde{E}_{\chi\gamma}^i. \end{aligned} \quad (3.17)$$

where $\tilde{E}_{\alpha\beta}^i$ is the traceless part of the strain-rate tensor [100], and λ^i (known as the flow-tumbling or flow-aligning parameter) models the extent to which the director field of species i explicitly orients with the strain axis of the fluid (the direction of the positive eigenvalue of $\tilde{E}_{\alpha\beta}^i$).

The system evolves towards the minimum of the free energy, given by the molecular field [27, 34] (Eq.(2.14))

$$H_{\alpha\beta}^i = -\frac{\partial \mathcal{F}}{\partial Q_{\alpha\beta}^i} + \frac{\delta_{\alpha\beta}}{2} \left(\frac{\partial \mathcal{F}}{\partial Q_{\chi\gamma}^i} \right) \delta_{\chi\gamma}. \quad (3.18)$$

The dynamics of the nematic tensor associated with fluid i is [55] (Eq.(2.12))

$$\partial_t Q_{\alpha\beta}^i + u_\chi^i \partial_\chi Q_{\alpha\beta}^i - S_{\alpha\beta}^i = \Gamma H_{\alpha\beta}^i. \quad (3.19)$$

The nematic field itself generates stresses, which drive flows in the fluid. This is modelled by a body force

$$F_\alpha^{Q,i} = \partial_\beta \Pi_{\alpha\beta}^i \quad (3.20)$$

where the stress tensor is the sum of the elastic and active stresses [26, 27] (Eqs.(2.15), (2.19))

$$\Pi_{\alpha\beta}^i = \Pi_{\alpha\beta}^{i,el} + \Pi_{\alpha\beta}^{i,act}, \quad (3.21)$$

$$\begin{aligned} \Pi_{\alpha\beta}^{i,el} = & 2\lambda^i(Q_{\alpha\beta}^i + \delta_{\alpha\beta}/2)Q_{\chi\gamma}^i H_{\chi\gamma}^i - \lambda^i H_{\alpha\chi}^i(Q_{\chi\beta}^i + \delta_{\chi\beta}/2) \\ & - \lambda^i(Q_{\alpha\chi}^i + \delta_{\alpha\chi}/2)H_{\chi\beta}^i - \partial_\alpha Q_{\chi\gamma}^i \frac{\partial \mathcal{F}^{LdG}}{\partial \partial_\beta Q_{\chi\gamma}^i} \\ & + Q_{\alpha\chi}^i H_{\chi\beta}^i - H_{\alpha\chi}^i Q_{\chi\beta}^i, \end{aligned} \quad (3.22)$$

$$\Pi_{\alpha\beta}^{i,act} = -\zeta_i \phi^i Q_{\alpha\beta}^i. \quad (3.23)$$

We emphasize here that the active stress generated by each component in Eq.(3.23) scales with the concentration fraction of the corresponding fluid ϕ^i i.e. the active force generated by component 1 is proportional to ϕ , while the active force generated by component 2 is proportional to $1 - \phi$.

Now, we return to the equation of motion for the first compressible fluid component. Using Eqs. (3.1) and (3.4), the momentum equation (3.2) for the first component can be written as

$$\begin{aligned} \rho^1[\partial_t u_\alpha^1 + u_\beta^1 \partial_\beta u_\alpha^1] = & -\phi \partial_\beta \sigma_{\alpha\beta}^{thermo} + F_\alpha^{visc,1} + F_\alpha^{body,1} \\ & -f\phi u_\alpha^1 + [\gamma\phi(u_\alpha^c - u_\alpha^1) + \phi(1-\phi)\partial_\alpha \delta\mu] \end{aligned} \quad (3.24)$$

where $\delta\mu = -\delta\mathcal{F}/\delta\phi$ [97, 98]. Defining an internal force density acting between the fluids,

$$G_\alpha = \gamma(u_\alpha^c - u_\alpha^1) + (1-\phi)\partial_\alpha \delta\mu, \quad (3.25)$$

Eq. (3.24) can then be rewritten as

$$\rho[\partial_t u_\alpha^1 + u_\beta^1 \partial_\beta u_\alpha^1] = -\partial_\beta \sigma_{\alpha\beta}^{thermo} + G_\alpha + \frac{F_\alpha^{visc,1} + F_\alpha^{body,1}}{\phi} - f u_\alpha^1. \quad (3.26)$$

The equation of the second compressible fluid component follows by symmetry.

In addition to solving the combined fluid using a normal LB algorithm, we also solve Eqs.(3.26) and (3.1) for the first fluid component using a modified LB scheme with two distribution functions, described in Refs. [97, 98], and also in Appendix A. We benchmarked the two-fluid code by comparing the LB results with a finite difference code, finding excellent agreement between the two alternate solvers.

In the limit $\gamma \rightarrow \infty$, the two component fluids are constrained to have the same velocity field, $u_\alpha^1 = u_\alpha^2 = u_\alpha^c$, and our model reduces to the commonly used single-fluid lyotropic model formalism [64].

Simulations are run in a periodic box of size 200×200 for 20,000 LB time-steps, with a random initial director configuration. In this chapter, we will consider a mixture of an active nematic and a passive isotropic fluid, hence the second nematic field is undefined ($\Gamma^2 = C^2 = \zeta^2 = K_{LC}^2 = S_{nem}^2 = \lambda^2 = L_{12} = 0$). The remaining parameter values are chosen as $\rho^C = 40$, $\gamma = 4$, $\nu^1 = \nu^2 = 1/6$, $a = 0.0025$, $\kappa = 5$, $\Gamma^1 = 0.1$, $C^1 = 0.1$, $\zeta^1 = 0.1$, $K_{LC}^1 = 0.15$, $S_{nem}^1 = 1$, $\lambda^1 = 0$ unless otherwise indicated. The system is initialized from a uniformly mixed configuration ($\rho^1 = 20$, $\rho^2 = 20$) with added noise in the configuration field, unless otherwise specified.

3.3 Phase separation driven by activity.

We solve the equations of motion for a mixture which comprises equal concentrations of an active nematic fluid and a passive fluid. The mixture is incompressible so that the total fluid density remains constant, but the relative concentrations of individual components can vary in space and time. The two components are initially homogeneously mixed so that the concentration of the active component $\phi = 0.5$ everywhere. However, above a threshold activity the system quickly self-organises into dynamically-evolving regions which are, respectively, rich or poor in the active fluid component. A typical snapshot of the phase-ordered state is shown in Fig. 3.2(a). The elongated domains have a higher concentration of active material, and continually break up and re-form. They are elongated by extensile active flows [64] and then pulled apart by the chaotic active turbulent flows.

We measure the standard deviation of ϕ , denoted by Δ , to quantify the degree of phase separation. Fig. 3.2(b) shows how Δ evolves with time towards a dynamical steady state.

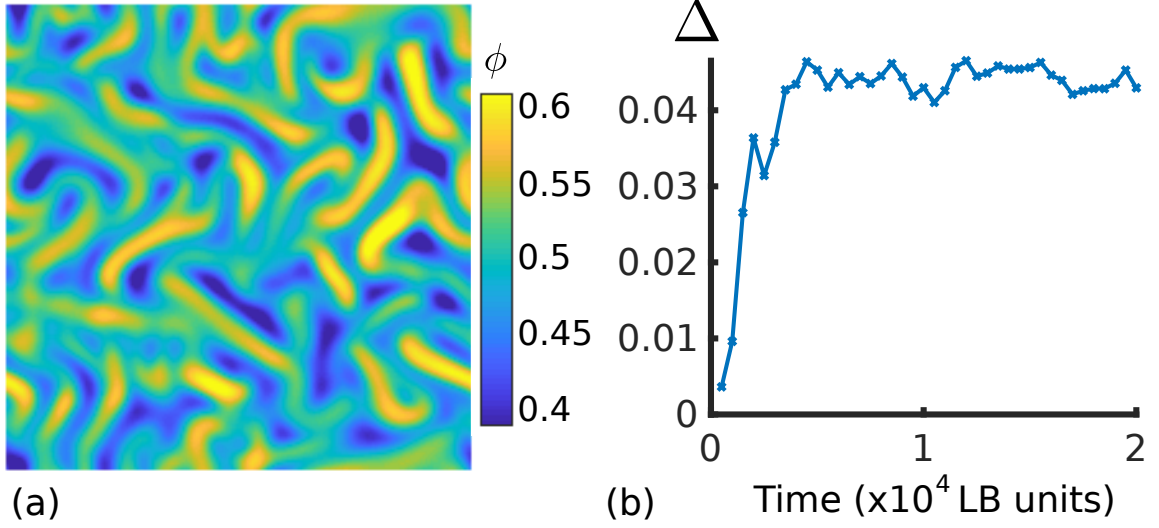


Figure 3.2: Active phase ordering: (a) Snapshot of the concentration profile in the micro-phase-separated state, color bar denotes concentration of the active phase (b) Standard deviation of the concentration, Δ , as a function of time.

3.3.1 Mechanism

We emphasise that the phase separation is entirely due to the activity, and requires no passive forces. The free energy in Eq. (3.9) is chosen to be a single well potential, $a > 0$, which favours mixing. To explain the mechanism causing the phase ordering consider a small fluctuation in concentration. From Eq. (3.21) this will lead to an active force tangential to the interface between the higher and lower activity regions

$$\mathbf{F}^{tangential} = 2\zeta|\nabla(S\phi)|(\mathbf{m} \cdot \mathbf{n})(\mathbf{l} \cdot \mathbf{n})\mathbf{l}, \quad (3.27)$$

where $\mathbf{m}$ and $\mathbf{l}$ are unit vectors normal (pointing away from the more active region) and tangential to the interface respectively, and $\mathbf{n}$ is a unit vector along the nematic director (Fig 3.3(a)) [64]. This force sets up flows parallel to the interface. The nematic director rotates in the flow leading to a net alignment parallel (perpendicular) to the interface in extensile (contractile) systems. This is active anchoring [64], as has been discussed in Sec. 2.5.2.

Similarly the active force normal to the interface is

$$\mathbf{F}^{normal} = -\zeta|\nabla(S\phi)|(2(\mathbf{m} \cdot \mathbf{n})^2 - 1)\mathbf{m}. \quad (3.28)$$

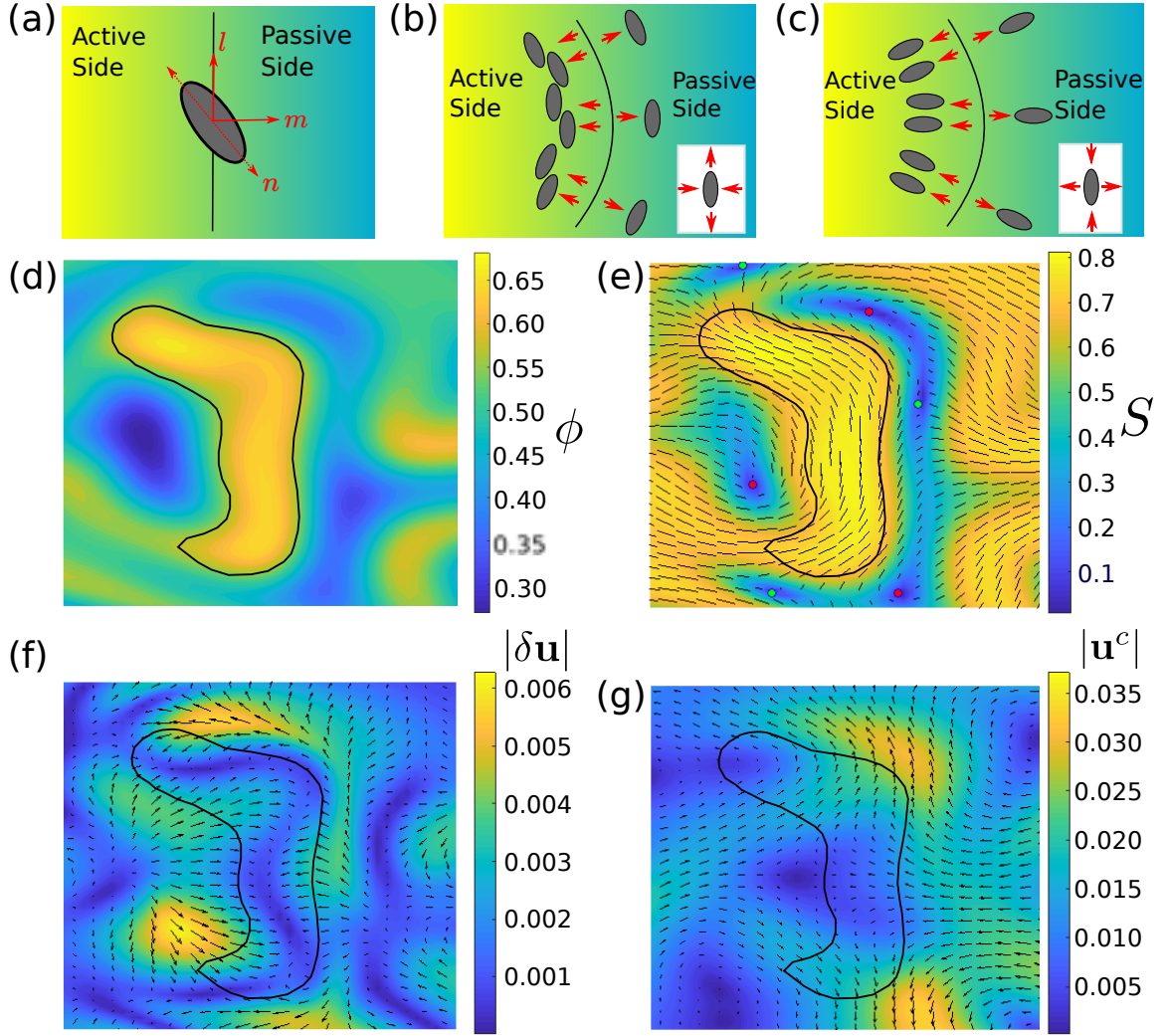


Figure 3.3: Active anchoring at an interface leads to a net flow normal to the interface: (a) Vectors introduced in the text. (b) Extensile nematics with planar active anchoring (inset: extensile flow field); (c) Contractile nematics with homeotropic active anchoring (inset: contractile flow field). In both cases the changing concentration of nematic particles leads to a net flow up the concentration gradient (red arrows). (d)-(g): Simulation snapshot of an actively phase-separated droplet. (d) Concentration field. (e) Director field, with planar active anchoring everywhere except at the tips. (f) Velocity difference between the active and passive fluid components, which points into the droplet except near $+1/2$ defects. (g) The combined flow field stretches the droplet, in a way identical to the single fluid result.

In the absence of anchoring, $\mathbf{F}^{normal}$ averages to zero. However, when there is active anchoring of the director, the force drives the active component up the concentration gradient towards the active nematic region. Hence the relative flow between the two components enhances concentration fluctuations to form active regions of concentration $\phi > 1/2$. The same mechanism leads to domain

formation in contractile systems, as the change in the direction of the active anchoring is compensated by the change in sign of the activity. This is illustrated schematically in Figs. 3.3(b),(c). A simulation snapshot of an active droplet is shown in Figs. 3.3 (d)-(g).

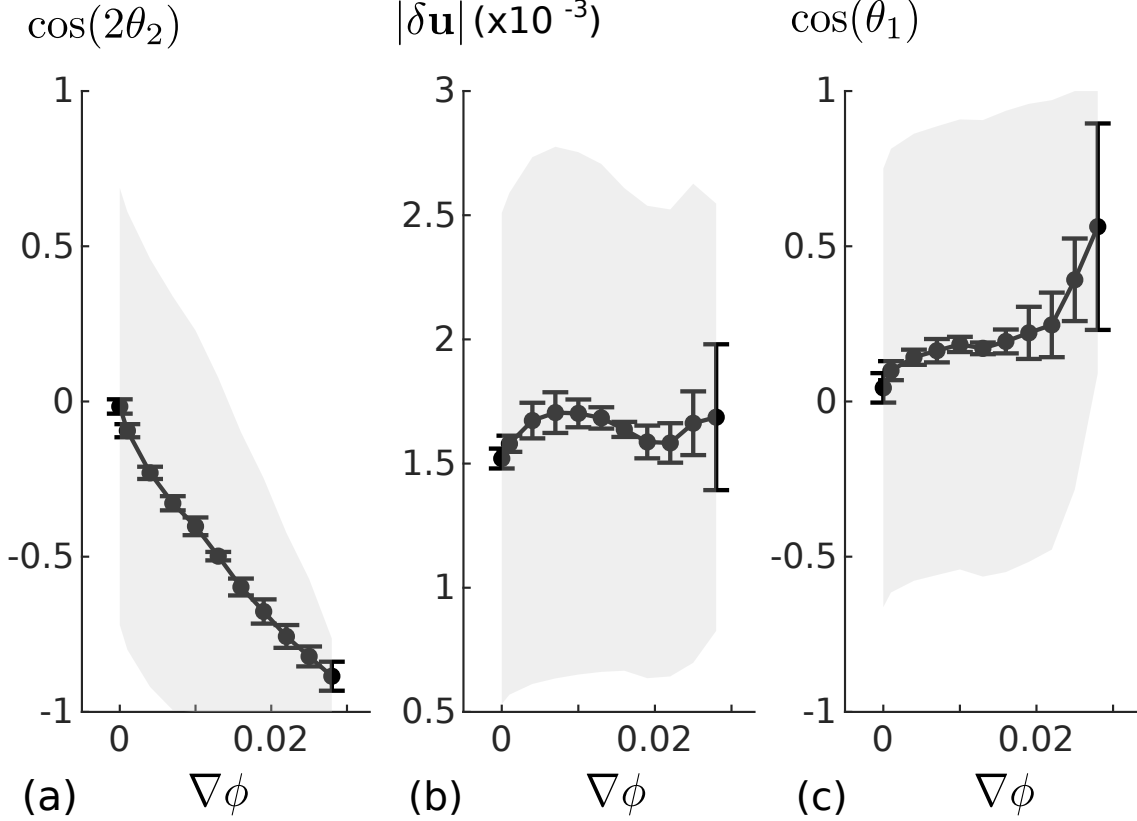


Figure 3.4: Flow–interface correlations: (a) Active anchoring increases with concentration gradients. (b) Magnitude of mean relative flow, $|\delta\mathbf{u}|$, does not depend on concentration gradients. (c) Orientation of mean slip velocity points up concentration gradients. Results are for extensile activity. Shaded regions show standard deviations of observed values.

To confirm the relevance of interfacial flows we numerically measured the dependence of physical quantities on the modulus of the concentration gradient, averaging over the whole domain. Projecting the nematic director field onto the concentration gradient yields $\cos(2\theta_2) = (\nabla\phi^T \cdot \mathbf{Q} \cdot \nabla\phi) / (S|\nabla\phi|^2)$, where θ_2 is the angle between the director field and $\nabla\phi$. This quantity tends to -1 for extensile activity confirming planar active anchoring (Fig. 3.4(a)). Similarly, it approaches +1 for contractile activity corresponding to homeotropic active

anchoring. The magnitude of the slip velocity $|\delta\mathbf{u}|$ is independent of the concentration gradient $|\nabla\phi|$ (Fig. 3.4(b)). However, the cosine of the angle between $\delta\mathbf{u}$ and $\nabla\phi$, $\cos(\theta_1) = (\nabla\phi \cdot \delta\mathbf{u})/(|\nabla\phi||\delta\mathbf{u}|)$, increases at large $|\nabla\phi|$, confirming that the direction of the relative fluid velocity is preferentially up concentration gradients (Fig. 3.4(c)).

These measured values have a large spread because the system is in a chaotic active turbulent state, however the mean values show the expected trends at interfaces consistent with the mechanism of phase separation outlined above.

3.4 Varying the Model Parameters

We now discuss more detailed numerical results investigating how the size of the phase separated domains and the difference in concentration between the two phases, Δ , depend on the model parameters. The mean of ϕ is fixed, since the total amount of active material is fixed. Therefore, we quantify the difference in concentration using the standard deviation of the concentration field at a given point in time, Δ . Fig. 3.5(a) shows that Δ increases, but does not scale linearly, with the activity coefficient ζ . This is because at higher activities the flow is more turbulent with many defects which reduce the strength of the active anchoring.

The formation of concentration gradients is opposed by the bulk free energy which scales as a , the relative drag γ , and the surface tension κ . We consider the balance between the activity and each of these in turn, choosing parameters where the given relaxation mechanism is dominant. Results are presented for extensile activity but they also hold for contractile systems since the same mechanisms are at play.

In Fig. 3.5(b) Δ is plotted as a function of $\zeta/a^{0.5}$ showing collapse onto a single curve. The collapse is a result of the balance between the driving force normal to the interface and the thermodynamic force which restores mixing. The former scales as ζ^2 because it depends on the strength of the anchoring and on the magnitude of the active force, both of which scale with ζ . The latter scales as a .

The data also lies on a single curve when Δ is plotted against the ratio ζ/γ (Fig. 3.5(c)). This shows the expected inverse relation between the activity, which creates a slipping velocity between the active and passive fluids, and the viscous

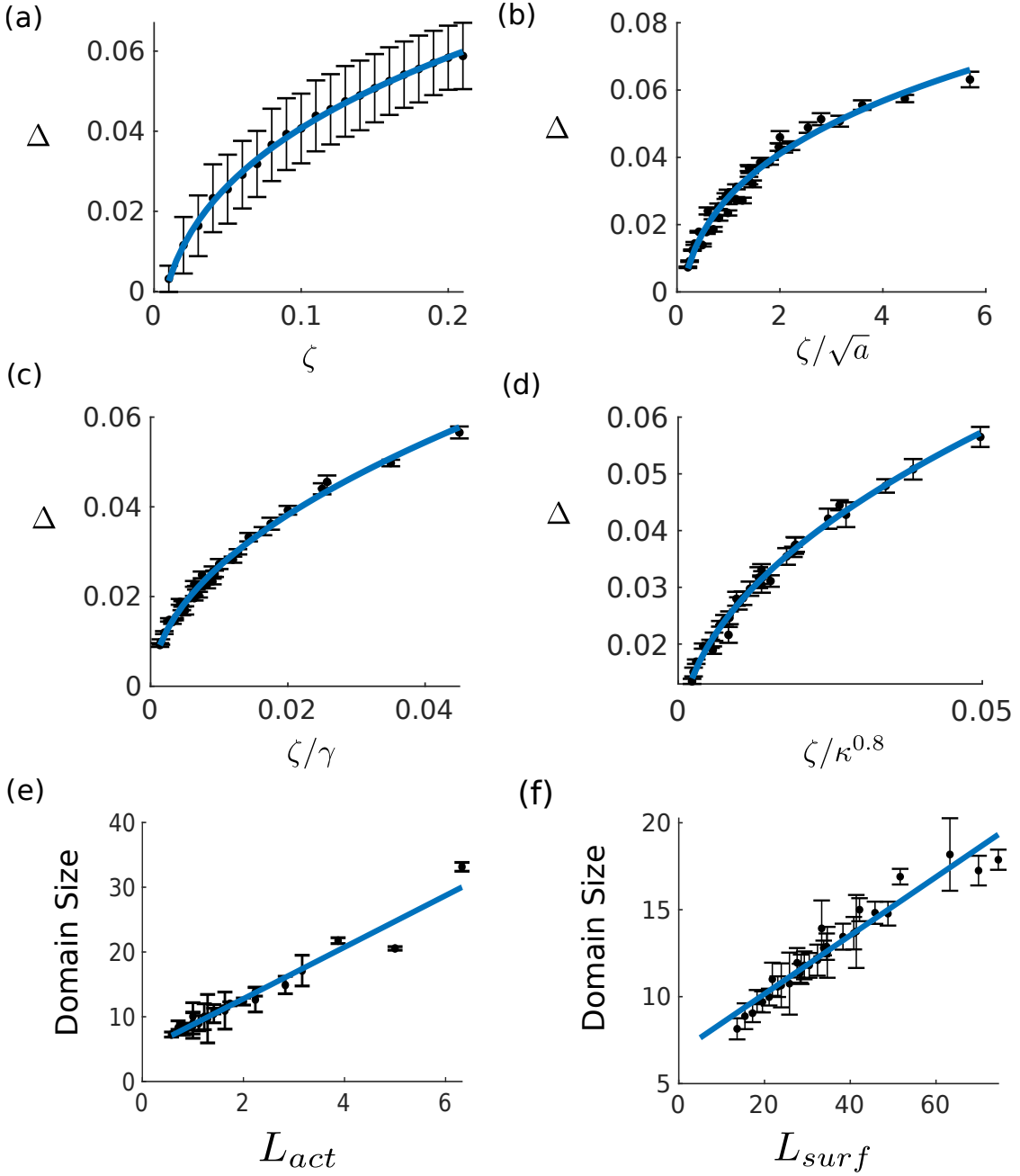


Figure 3.5: Variation of concentration difference Δ and average domain size with model parameters: In all the plots above, we vary activity $\zeta \in [0.03, 0.18]$. (a) Δ scales non-linearly on increasing activity. Ordering is primarily opposed by (b) bulk free energy $a \in [0.0010, 0.0075]$, (c) drag between the fluids $\gamma \in [4, 22]$, (d) surface tension $\kappa \in [5, 25]$. Average domain size as a function of the (e) active length-scale, L_{act} , for $K_{LC} \in [0.05, 0.40]$. (f) Surface tension length-scale, L_{surf} , for $\kappa \in [5, 25]$. Blue curves are best-fit power laws for (a)-(d), and best-fit straight lines for (e), (f). Error bars show standard deviations from 40 simulation values.

drag that dampens the slip. We expect to see no phase separation in the limit $\gamma \rightarrow 0$, corresponding to no relative flows between the component fluids.

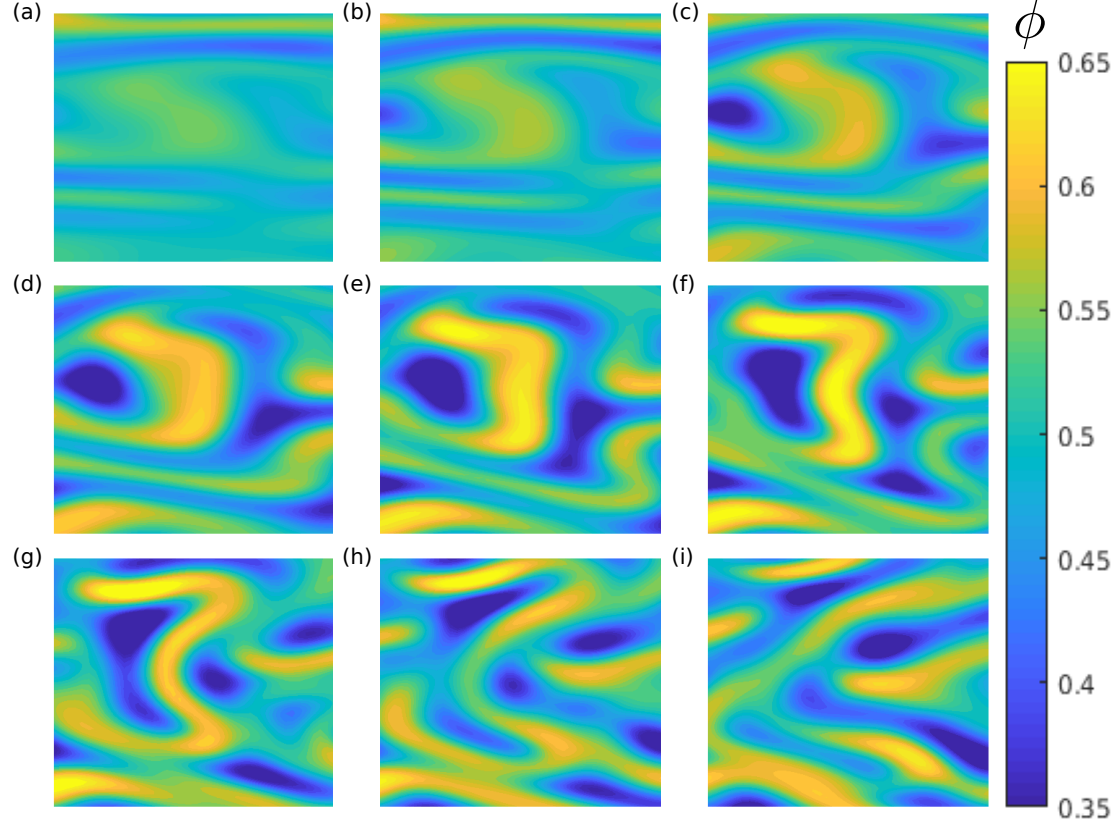


Figure 3.6: Formation and dissolution of an extensile active droplet. Snapshots are taken at 200 time step intervals. (a)-(c): A small concentration fluctuation develops and increases with time, forming a new active droplet. (d)-(f): The droplet grows in size and starts to form a bend instability. (g)-(i): The bend instability gets larger, and splits the droplet apart into multiple smaller droplets. Colour bar denotes the local concentration of the active component.

A similar plot, obtained by changing the surface tension coefficient κ at constant a and γ , shows the best data collapse when Δ is plotted against $\zeta/\kappa^{0.8}$ (Fig. 3.5(d)). We do not have a simple argument for this dependence as several factors contribute. In addition to the thermodynamic penalty, an increase in surface tension weakens the activity gradient by making the interface more diffuse, and hence will alter the details of the active interfacial forces and active anchoring.

The length-scale of the domains can be calculated numerically from the first zero of the spatial correlation function of the fluctuations in the concentration

field. For fixed surface tension, Fig. 3.5e shows that the domain size is controlled by the usual active nematic length scale $L_{act} = \sqrt{K_{LC}/\zeta}$. This is, however, not the only length scale controlling the physics. The domain size can be changed by varying a second length scale, related to the surface tension, $L_{surf} = \sqrt{\kappa\eta/\zeta}$, as shown in Fig. 3.5f. Active droplets break up by forming bend or splay instabilities with typical lengthscale L_{act} , or by self-shearing instabilities [64, 101] with typical lengthscale L_{surf} . Snapshots showing formation and dissolution of a droplet are shown in Fig. 3.6.

3.5 Threshold for phase ordering

It is possible to obtain an exact expression for the activity threshold above which phase ordering occurs, in the limit of perfect surface anchoring, and ignoring interface curvature. We discuss the effects of curvature in Sec. 3.6.

Consider a concentration gradient around $\phi = 1/2$ in the x -direction, given by $\nabla\phi = |\nabla\phi| \hat{x}$. Let the activity coefficient be $\pm|\zeta|$, where the $+$ ($-$) sign denotes extensile (contractile) activity. Furthermore, in this section, we choose a functional form of S_{nem} that depends on the local concentration of nematogens $S = S_{nem}\sqrt{\phi}$. This choice modifies the numerical prefactors for the active force at an interface below, but does not make a qualitative difference to the mechanism or direction of active forces at an interface. For perfect active anchoring at the interface, this leads to a director field $Q_{xx} = \mp S_{nem}\sqrt{\phi}$, $Q_{xy} = 0$.

The normal active force at the interface is then $\mathbf{F}_{act} = (3/2)S_{nem}\sqrt{\phi} |\zeta| |\nabla\phi| \hat{x}$, and the restoring force driven by the free energy is $\mathbf{F}_{rest} = -2a\rho^c\phi(1-\phi)|\nabla\phi| \hat{x}$. This leads to an expression for the slip velocity δu_x in the Stokes limit:

$$\delta u_x = \frac{F_{act} + F_{rest}}{\gamma\phi(1-\phi)} = \frac{(3/2) |\zeta| S_{nem}\sqrt{\phi} - 2a\rho^c\phi(1-\phi)}{\gamma\phi(1-\phi)} |\nabla\phi| \hat{x}. \quad (3.29)$$

The concentration gradient will increase for activities greater than

$$|\zeta|_{crit} = \frac{4a\rho^c\phi(1-\phi)}{3\sqrt{\phi}S_{nem}}. \quad (3.30)$$

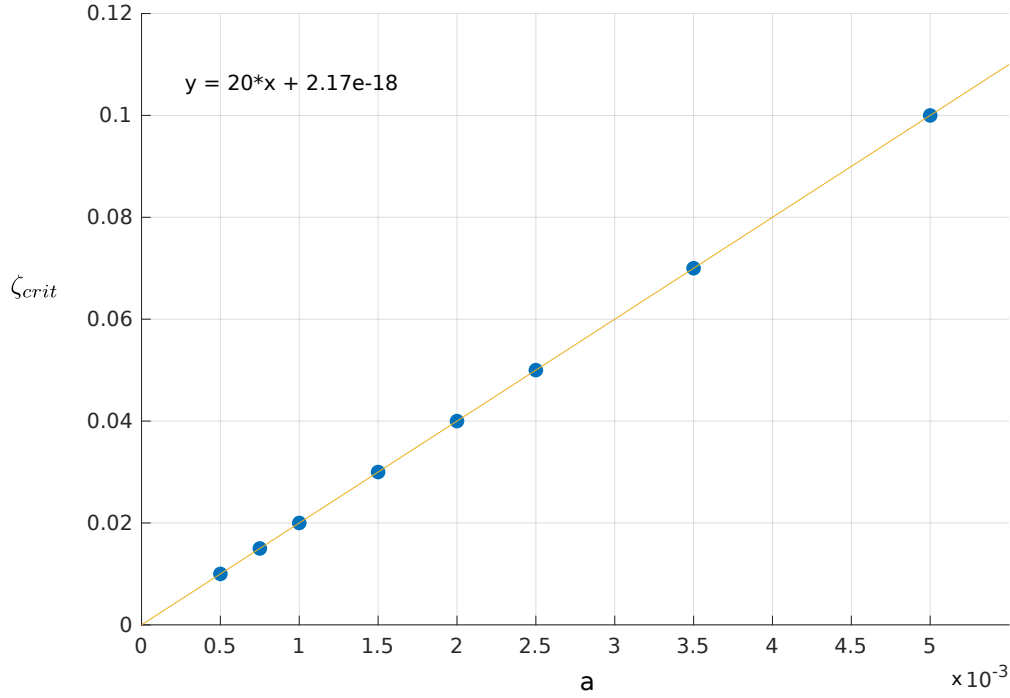


Figure 3.7: Critical activity when initialized in an aligned orientational state with small concentration fluctuations around $\phi = 1/2$ ($\delta\phi \approx 10^{-3}$). We evaluate Δ at a later point in time (2000 LB time-steps), and the critical activity is the lowest value of activity where Δ is larger than the starting value. The yellow line is $y = 20x$. The critical activity is measured to be linear in a as predicted. The slope is 20.0 compared to the theoretical estimate 18.9. The difference is likely to be due to neglecting the viscous dissipation in the fluids in the theoretical estimate.

In the simulations reported in the chapter, the critical activity for phase separation is, in fact, slightly lower than the critical value in Eq.(3.30) verified above because the initial conditions we use lead to fluctuations in the orientational field. Bend and splay instabilities form, and these have regions with concave distortions which lower the critical activity, as discussed in Sec 3.6.

We can verify the critical activity numerically by studying an aligned orientational state with small concentration fluctuations around $\phi = 1/2$ ($\delta\phi \approx 10^{-3}$). The concentration fluctuations are magnified if the activity is above the critical activity, but dies out for activities below the threshold. According to Eq.(3.30), we expect to see a linear relation between $|\zeta|_{crit}$ and a . This is shown in Fig. 3.7. Plugging in numbers, we expect the slope to be 18.9 but we find the slope to be 20 instead - this is likely due to neglecting viscous dissipation in the theoretical estimate.

3.6 Effect of Distortions in the Director Field

The usual active stresses arising from distortions in the nematic field can either help or hinder the formation of the instability depending on the curvature of the interface.

To estimate the magnitude of this effect we consider a circular surface centred at the origin of a polar coordinate system with radius R , and with the director field anchored at the surface with an orientation $\Theta(\theta)$. We also assume that there is a gradient of nematic order S and concentration field ϕ across the boundary, with variations only in the radial direction $S(r)\phi(r)$.

Exploiting symmetries of the nematic tensor, the active force $\mathbf{F} = -\nabla \cdot (\zeta\phi\mathbf{Q})$ is given by

$$F_x = -\zeta \left(\partial_x(\phi Q_{xx}) + \partial_y(\phi Q_{yx}) \right), \quad (3.31)$$

$$F_y = -\zeta \left(\partial_x(\phi Q_{yx}) - \partial_y(\phi Q_{xx}) \right). \quad (3.32)$$

Using

$$\begin{aligned} Q_{xx} &= S(r) \cos(2\Theta(\theta)), & Q_{yx} &= S(r) \sin(2\Theta(\theta)), \\ \partial_x &= \cos\theta \partial_r - \frac{\sin\theta}{r} \partial_\theta, & \partial_y &= \sin\theta \partial_r + \frac{\cos\theta}{r} \partial_\theta, \end{aligned}$$

we find

$$F_x = -\zeta \cos(2\Theta - \theta) \left(\frac{\partial(\phi S)}{\partial r} + \frac{2S}{R} \frac{\partial\Theta}{\partial\theta} \right), \quad (3.33)$$

$$F_y = -\zeta \sin(2\Theta - \theta) \left(\frac{\partial(\phi S)}{\partial r} + \frac{2S}{R} \frac{\partial\Theta}{\partial\theta} \right). \quad (3.34)$$

For strong homeotropic anchoring in contractile ($\zeta < 0$) systems, $\Theta = \theta$, and in radial coordinates, Eqns. (3.33) and (3.34) reduce to

$$F_r = -\zeta \left(\frac{\partial(\phi S)}{\partial r} + \frac{2S}{R} \right), \quad (3.35)$$

$$F_\theta = 0. \quad (3.36)$$

For strong planar anchoring in extensile ($\zeta > 0$) systems, $\Theta = \theta + \pi/2$, and

$$F_r = \zeta \left(\frac{\partial(\phi S)}{\partial r} + \frac{2S}{R} \right), \quad (3.37)$$

$$F_\theta = 0. \quad (3.38)$$

Note that $\frac{\partial(\phi S)}{\partial r} < 0$, since the concentration and ordering of the active material are higher inside the droplet. This term drives the phase separation. The second term results from gradients in the director field because of the anchoring at a curved interface. If the interface is convex this is a radial force opposing phase separation for both extensile and contractile activity. This force decreases with increasing droplet radius and, in general, a circular droplet is stable if the drop radius is a few times the interface width. Using typical values from our simulations the two forces balance for $R \sim 7$ LB units.

Note, however, that in the simulations drops are elongated so much of their surface is flat and the effect of director distortions is reduced. Moreover, in Fig. 3.3 (e), we observe that the ends of the droplets are not actively anchored because of the high elastic energy cost at a sharply curved interface. These effects both help to stabilize the droplets.

If the surface is concave and actively anchored, the radial flow induced by the orientation field now supports the flow driving phase separation. Also, if the surface is anchored in the opposite orientation to active anchoring by passive forces, distortions once again generate force enhancing droplet formation.

3.7 Summary

By introducing an active two-fluid model with viscous drag between the fluids we have found that a homogeneous mixture of an active nematic fluid and a passive fluid spontaneously phase orders into regions with different concentrations of each fluid component. This is driven dynamically by active anchoring and normal active forces at concentration gradients, and is an example of liquid-liquid phase separation [72] out of thermodynamic equilibrium.

The two-fluid model reduces to the lyotropic formulation of nemato-hydrodynamics that has been successful in defining active and passive regions within a simulation, in the limit of infinite viscous drag between the fluids. However, because the lyotropic approach uses a single velocity field for both phases [64, 102], it cannot account for relative flows between, or compressibility of, the two components.

In this chapter, we have also seen that the magnitude of phase separation and the resultant domain sizes are dictated by the balance between activity, surface tension, and free energy. We then theoretically derived the minimum activity required for phase separation, and discussed the effect of system parameters and interface curvature on these results.

In the next chapter, we will extend these results to the case where both species are active nematics with different activities.

Take-home message:

An active nematic fluid mixed with a passive isotropic fluid spontaneously phase-separates into turbulent micro-phases.

“Algebra’s like sheet music, the important thing isn’t ‘can you read music’, it’s ‘can you hear it’. Can you hear the music, Robert?”

Neils Bohr to Robert Oppenheimer
(“Oppenheimer”, 2023)

4

Phase Ordering in Binary Mixtures of Active Nematic Fluids

In this chapter, we extend the results of the previous chapter to discuss mixtures of two active nematics, each with different activities, coupled by viscous drag. This is motivated by recent experiments which demonstrate cell sorting in mixtures of extensile and contractile cells which have different activities [103, 104].

The rest of this chapter is organized as follows: In Sec. 4.1, we discuss the angle between the orientations of the two active species when they are coupled only by viscous drag. In the absence of flow alignment, we find that species with the same sign of activity align parallel to each other, while species with opposite signs of activity align perpendicular to each other. On increasing the flow-alignment, the species tend to align parallel with each other regardless of activity. In Sec. 4.2, we move on discuss phase separation in active-active mixtures. We review the mechanism discussed in Sec. 3.3.1, and show how this is modified when both species are active. We also study how varying the activity of each species affects the magnitude of phase separation, with and without elastic coupling between nematogens. In Sec. 4.3, we discuss how changing concentration fractions and friction affect the magnitude of phase separation in extensile-contractile mixtures,

and how this changes the resultant morphology. Finally, in Sec. 4.4, we conclude by summarizing the results from this chapter.

For the rest of this chapter, we use the parameters $\rho^C = 40$, $\gamma = 4$, $\kappa = 5$, $a = 0.0001$, $b = 0.0001$, $\eta^1 = \eta^2 = 10/3$, $S_{nem}^1 = S_{nem}^2 = 1$, $C^1 = C^2 = K^1 = K^2 = 0.1$, $L_{12} = 0.1$, $\lambda = 0$, $f = 0$, and $\Gamma^1 = \Gamma^2 = 0.1$, unless otherwise specified. ζ_1, ζ_2 are varied in the range $[-0.1, 0.1]$. We simulate the equations for 50,000 time steps on a 200 x 200 grid with periodic boundary conditions. The initial condition was chosen to be $\rho^1 = \rho^2 = 20$, with the initial director and velocity field configurations obtained by simulating 500 LB time-steps from a randomly initialized director configuration without active forcing.

4.1 Active flows introduce director alignment between nematic fluid components

In this section, we argue that there is parallel or perpendicular alignment between the two components of the nematic mixture due to the director fields aligning with the rate of strain tensor in the combined fluid. Throughout this section, we set $L_{12} = 0$ and $a = 0.2$ to study flow-induced director-director coupling in the absence of imposed elastic alignment or phase separation. We first consider the flow aligning parameter $\lambda^1 = \lambda^2 = \lambda = 0$, and then discuss the effects of a non-zero λ .

4.1.1 Zero flow-aligning parameter ($\lambda = 0$)

Assuming that the momentum balance in each fluid (Eq. (3.2)) is dominated by viscous and active stresses then, for $\lambda = 0$, Eqs. (3.12) and (3.23) give a force balance

$$\partial_\beta \eta^i E_{\alpha\beta}^c = \zeta_i \partial_\beta Q_{\alpha\beta}^i \quad (4.1)$$

for each of the fluid components. Here, we have assumed that both component fluids have approximately the same strain rate $E_{\alpha\beta}^c$.

If $\zeta_i > 0$ (extensile), $Q_{\alpha\beta}$ has the same sign as $E_{\alpha\beta}$, and the nematic orientation $\theta_{director}$ aligns along the stretching direction of the combined fluid θ_{strain} ($\theta_{director} \parallel \theta_{strain}$). θ_{strain} is the direction corresponding to the (largest) positive

eigenvalue of the matrix $E_{\alpha\beta}$. If $\zeta_i < 0$ (contractile), $Q_{\alpha\beta}$ and $E_{\alpha\beta}$ have opposite signs, and the nematic aligns along the compression direction of the combined fluid ($\theta_{director} \perp \theta_{strain}$). This is shown in Fig. 4.1(a). The spread in the distribution is due to elasticity in the nematic field of each component and the effect of the passive backflow terms in the Navier Stokes equations which are neglected in Eq. (4.1).

When two active fluids are strongly coupled through viscous drag, the alignment of each nematic with the combined fluid velocity gradient induces an effective alignment between the two director fields. Both species orient in the same (perpendicular) direction if they have the same (opposite) sign of activity. The overall degree of alignment can be characterised by calculating the average value of $\cos(2(\theta_1 - \theta_2))$, where θ_i is the angle of the i -th director field. This quantity is +1 for parallel alignment and -1 for perpendicular alignment as shown in Fig. 4.1(b).

The alignment of the director fields increases with increasing the activity of the fluids. The alignment is strongest when both fluids have the same magnitude of activity (see Figs. 4.1(b) and 4.1(d)).

4.1.2 Effect of the flow-aligning parameter λ

We now consider the effects of having a large flow-aligning parameter $\lambda^1 = \lambda^2 = \lambda$. The flow-alignment term in Eq. (3.19) tends to align the nematic director field with the extensional strain axis, regardless of activity. For an extensile fluid this merely slightly enhances the alignment along the strain axis caused by the activity. However, the angle between contractile nematogens and the strain axis forms a bimodal distribution with some regions aligning parallel to the strain, due to flow alignment, and others perpendicular to the strain, due to active flows, with the relative fraction of each alignment dependent on λ . (Fig. 4.1(c)). This is consistent with force balance (Eq. (3.2)), since there are now both flow-aligning and elastic terms to account for.

The flow alignment can, in turn, affect the effective alignment between the director fields of the two components. For extensile-extensile mixtures, there is just

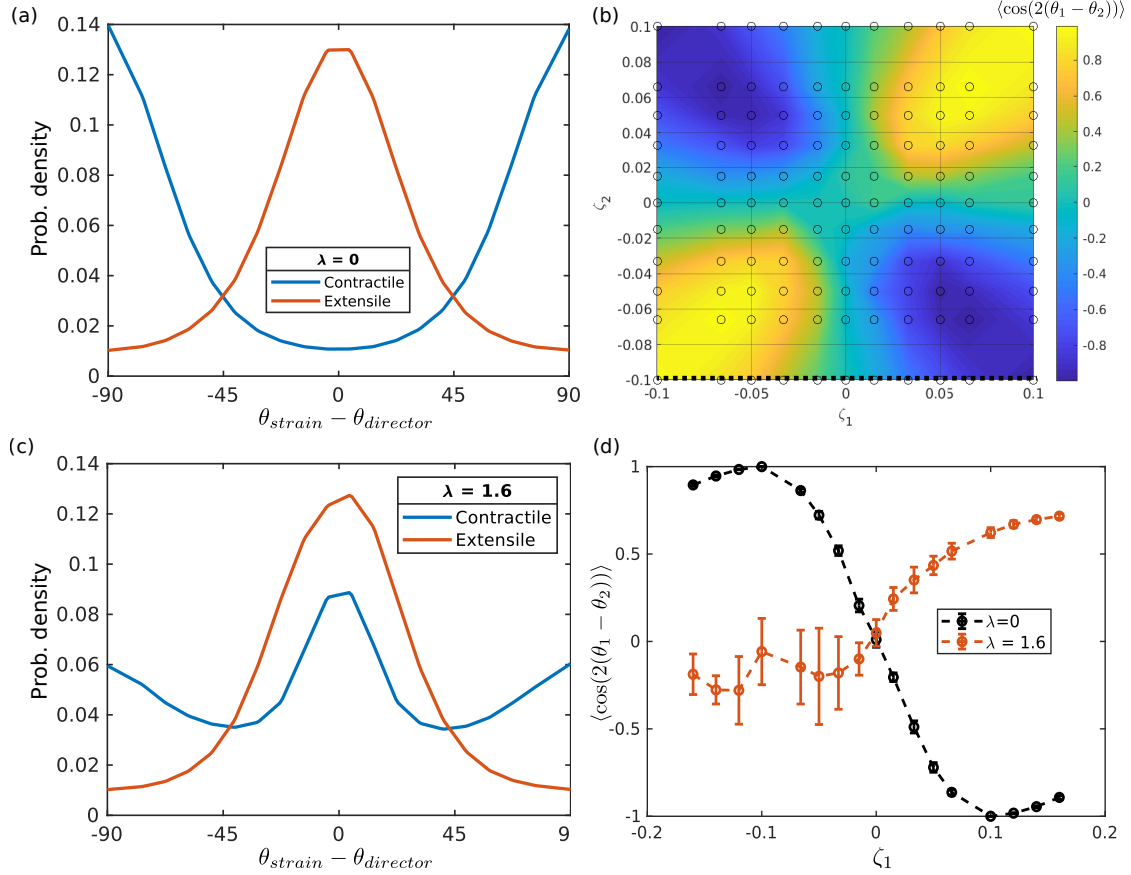


Figure 4.1: Alignment between nematic director fields. (a) Probability density function shows that for $\lambda = 0$, contractile nematics tend to align perpendicular to the extensional strain axis (blue), while extensile nematics align parallel (red) ($\zeta_1 = -0.10, \zeta_2 = 0.067$). (b) Contour plot showing nematic alignment between the different components for $\lambda = 0$. The colourbar shows $\langle \cos(2(\theta_1 - \theta_2)) \rangle$ which is +1 (yellow) for parallel and -1 (blue) for perpendicular. Circles denote individual simulations, while the contour plot shows the interpolated values. (c) Probability density function shows that for $\lambda = 1.6$, contractile nematics have a bimodal distribution preferentially aligning either parallel and perpendicular to the extensional strain axis (blue), while extensile nematics tend to align parallel (red) ($\zeta_1 = -0.10, \zeta_2 = 0.05$). (d) Cross-section of nematic alignment while varying ζ_1 , with $\zeta_2 = -0.10$. The $\lambda = 0$ cross-section is marked by the black dotted line in the contour plot. Changing to $\lambda = 1.6$ changes the nematic alignment significantly in both contractile-contractile ($\zeta_1 < 0$) and contractile-extensile ($\zeta_1 > 0$) mixtures.

a small decrease in $\langle\theta_1 - \theta_2\rangle$. For an extensile-contractile mixture, some contractile directors align with the extensional strain axis, leading to an increase in alignment between the two species. For a contractile-contractile mixture, both director fields are frustrated, and $\langle\theta_1 - \theta_2\rangle$ depends on the fraction of director fields which align parallel or perpendicular to the strain. In Fig. 4.1(d), we plot the average nematic alignment $\langle\cos(2(\theta_1 - \theta_2))\rangle$ along a cross-section varying ζ_1 keeping $\zeta_2 = -0.10$ fixed, for both $\lambda = 0$ (flow tumbling regime) and $\lambda = 1.6$ (flow aligning regime).

4.2 Phase Separation in Active-Active Mixtures

In this section, we first review the phase-separation mechanism and then apply it to the case of two active nematic fluids of different activities. Secondly, we determine how the strength of phase separation depends on the activities of each species. Finally, we look at how the results change when the nematic species are constrained to align with each other through elastic interactions.

4.2.1 Mechanism

Active phase separation begins when the two component species generate a flow fluctuation which locally moves them in different directions, setting up a small concentration gradient in the fluid. This difference in concentration leads to an imbalance in active stress across the interface, which drives active flows normal and tangential to the interface in each fluid.

We numerically verify this mechanism by looking at the active anchoring and flow alignment at concentration gradients (see Fig. 4.3 for typical values of concentration ϕ and the concentration gradient $|\nabla\phi|$ across an interface). We define the director angle of species i as θ_i , the orientation of the relative flow velocity $\delta\mathbf{u}$ as θ_v , and the direction of the gradient of ϕ as θ_ϕ . θ_ϕ is normal to the interface and points towards increasing ϕ . We quantify anchoring by measuring $\langle\cos 2(\theta_i - \theta_\phi)\rangle$, the angle between the director field and concentration gradient, which ranges from +1 for homeotropic anchoring to -1 for planar anchoring. At interfaces, the contractile fluid tends to align normal to the interface (Fig. 4.2(d)) while the extensile fluid prefers to align

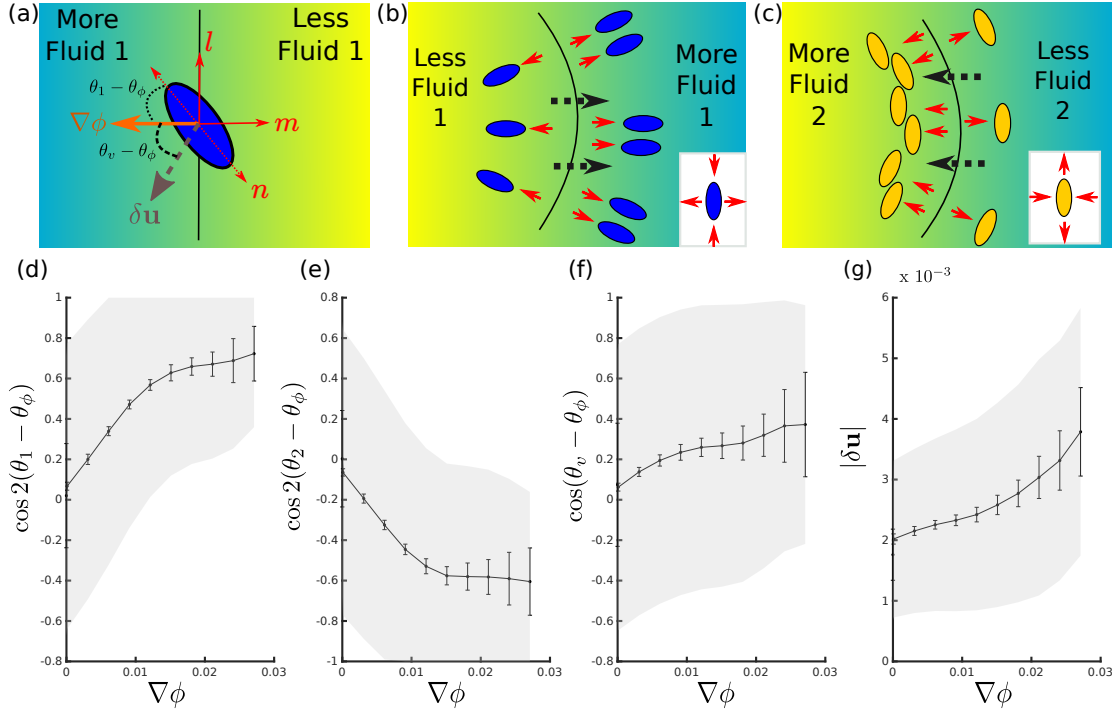


Figure 4.2: Mechanism of Phase Separation: (a) Interface normal $\mathbf{m}$, tangent $\mathbf{l}$, and nematic director $\mathbf{n}$ at an interface. Schematic representation of the mechanism of phase separation for (b) a contractile component 1 (blue) and (c) an extensile component 2 (yellow). Red arrows show the forces at the interface created by each nematogen, and black arrows show the net direction of the active force. (d)–(g) simulation data demonstrating the mechanism for $\zeta_1 = -0.10, \zeta_2 = 0.067$, and $L_{12} = 0$: (d) Contractile species align homeotropically at interfaces. (e) Extensile species align parallel to the interface. (f) The relative flow between the fluid components tends to orient normal to the interface, in the direction of increasing concentration gradient. (g) Relative flows between the fluids are stronger at the interface. The trend lines show the mean, combining data from 50 measurements taken in intervals of 1000 timesteps. The error bars indicate the standard deviation of the mean. The shaded region shows the standard deviation in the spread of the measured quantities.

tangentially (Fig. 4.2(e)). We also check the orientation of the relative flow $\delta \mathbf{u}$ with respect to the normal to the interface by measuring $\langle \cos(\theta_v - \theta_\phi) \rangle$, which is +1 for relative flows pointing towards higher ϕ , and -1 for relative flows towards lower ϕ . Figure 4.2(f) shows that the net flow between the fluid components tends to orient normal to the interface, in the direction of increasing concentration gradient. Finally Fig. 4.2(g) confirms that the magnitude of the relative flow $\langle |\delta \mathbf{u}| \rangle$ is stronger at interfaces. The same mechanism holds for imposed alignment between the nematogens, $L_{12} \neq 0$.

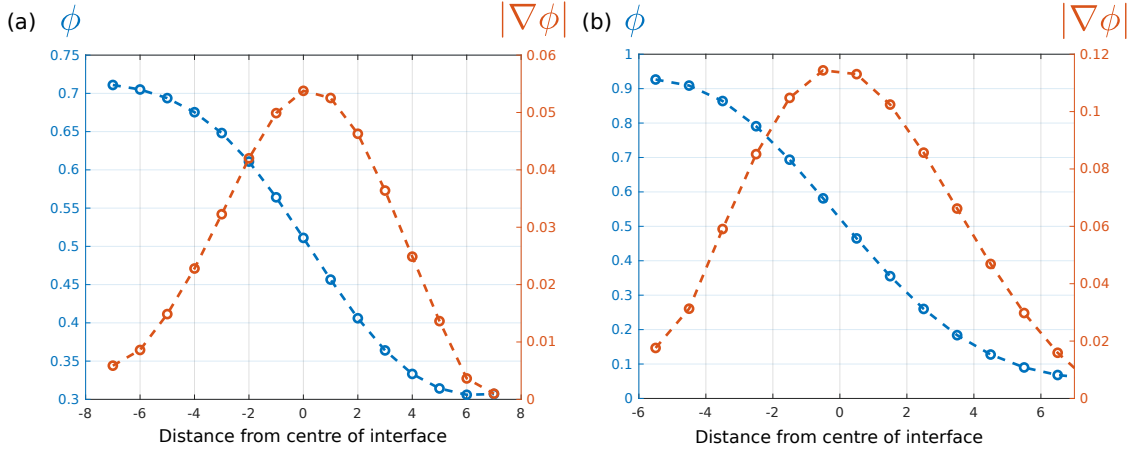


Figure 4.3: Cross-section of ϕ and $\nabla\phi$ across an interface. Snapshot of a typical interface cross-section with parameters for a mixture of two active nematics **(a)** with, and **(b)** without elastic director alignment. Blue curves show the concentration of the species 1, ϕ , and red curves show the magnitude of the concentration gradient, $|\nabla\phi|$. Distances on the x -axis are in LB units. The units of $\nabla\phi$ are inverse units of length, in Lattice-Boltzmann (LB) units. Since the interfaces are smooth and diffuse, the magnitude $|\nabla\phi|$ depends both on the difference in concentration between two neighbouring phases, as well as the typical lengthscale of the interface.

4.2.2 No imposed alignment between directors

In this subsection, we look at phase separation when there is no imposed elastic alignment between the nematic director fields of the two components ($L_{12} = 0$). Viscous drag between the fluids aligns the director fields parallel (perpendicular) to each other if the two species have the same (opposite) sign of activity. In all cases, we observe chaotic turbulent microphase separation, with phases forming and dissociating rapidly. A snapshot of the concentration field is shown in Fig. 4.4(a).

We characterize the magnitude of phase separation by calculating Δ , the standard deviation of the concentration field, which quantifies the variation from the uniformly mixed state. For a mixture with equal concentration fraction of each fluid component, Δ varies between 0 (no ordering) and $1/2$ (complete phase separation). A contour plot showing how Δ depends on the activities ζ_1 and ζ_2 is shown in Fig. 4.4(b). The magnitude of phase separation depends on the difference in magnitude of the activities, but not on their signs. This is because, although changing the sign of activity changes the relative alignment of the directors, the flow fields in both fluids remain in the same direction. Thus the phase separation is most pronounced

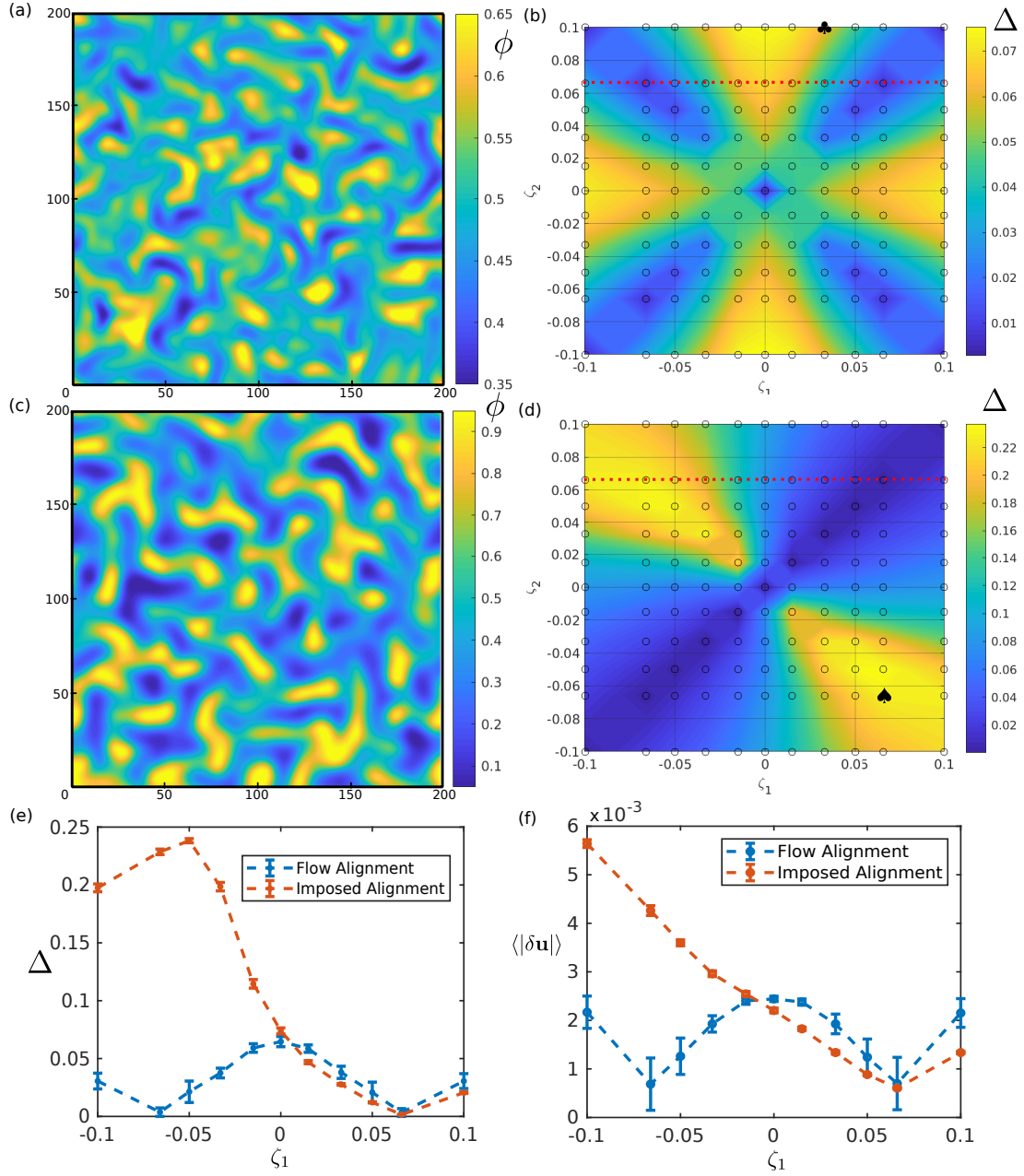


Figure 4.4: Phase separation in active-active mixtures: (a) Snapshot of the concentration field for a turbulent microphase separated state without imposed parallel alignment, $L_{12} = 0$ (corresponding to the point marked by ♣ in panel (b)). The colourbar shows local concentration ϕ . (b) Magnitude of the phase separation, Δ , for different activities. Circles denote individual simulations, while the contour plot shows the interpolated values. (c) Snapshot of the concentration field for a turbulent microphase separated state with imposed parallel alignment, $L_{12} = 0.1$ (corresponding to the point marked by ♠ in panel (d)). (d) Magnitude of the phase separation, Δ , for different activities. Circles denote individual simulations, while the contour plot shows the interpolated values. Note the increase in the magnitude of phase separation when there is elastic alignment between the director fields of the fluid components. Variation of (e) the magnitude of phase separation, Δ , and (f) the relative flow magnitude, $|\delta \mathbf{u}|$, with ζ_1 for $\zeta_2 = 0.067$ (i.e. along the red dotted lines in (b) and (d)), comparing $L_{12} = 0$ and $L_{12} \neq 0$.

when an active species is mixed with a passive one, corresponding to the maximum relative flows between the two fluids.

4.2.3 Imposing director alignment

We next impose $L_{12} \neq 0$, so that the director fields of the two nematic components are strongly aligned. A contour plot showing the magnitude of phase separation on varying the activities ζ_1 and ζ_2 is shown in Fig. 4.4(d).

If both species have the same sign of activity, the imposed parallel alignment merely reinforces the flow-induced alignment discussed in Sec. 4.1. The resultant phases look very similar to Fig. 4.4(a), but the magnitude of phase separation is slightly lower because the active flows are better aligned.

However, the magnitude of the phase separation is significantly higher if each fluid components have opposite signs of activity. This result in the forced parallel alignment case ($L_{12} \gg 0$) is drastically different than the case where the directors are free to align with the flow ($L_{12} = 0$, Sec. 4.2.2). The relative flow between the two components is much higher in the extensile-contractile case because each fluid generates active forces in opposite directions at a concentration gradient. The strongest phase separation, corresponding to the largest spread of concentration $\max(\phi) - \min(\phi) \approx 1$, is observed when a highly extensile fluid is mixed with a highly contractile one. The fluids form elongated droplet networks with large differences in concentration between the different regions, as shown in Fig. 4.4(c).

To directly compare the flow-alignment and imposed parallel alignment cases the variation of the magnitudes of phase separation, Δ , and the relative flow between the fluid components, $\delta \mathbf{u}$, with ζ_1 for a fixed ζ_2 are plotted in Fig. 4.4(e) and (f).

The requirement for strong phase separation in the extensile-contractile regime is not the exact form of the free energy, but that the director fields of the two species be aligned with each other. Instead of using an elastic cross-alignment term, a large flow-aligning parameter ($L_{12} = 0, \lambda \gg 0$) can also align extensile and contractile director fields (see Sec. 4.1). Increasing the flow-aligning parameter therefore

increases phase separation in an extensile-contractile mixture by improving director-director alignment, as shown in Fig. 4.5.

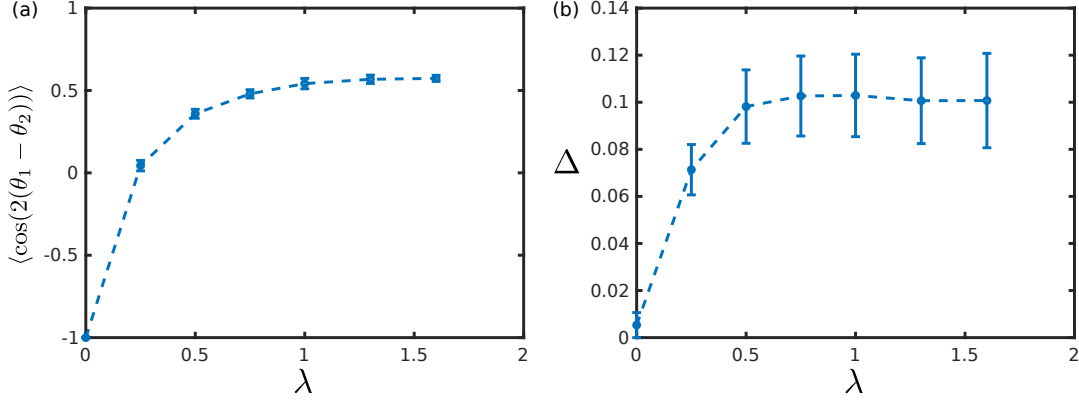


Figure 4.5: Effect of the flow-aligning parameter: We vary the flow-aligning parameter $\lambda^1 = \lambda^2 = \lambda$ for an extensile-contractile mixture ($\zeta_1 = 0.1, \zeta_2 = -0.1$), without imposed elastic director alignment. $\lambda = 0.5$ marks the transition from the flow-aligning to the flow-aligning regime. (a) Degree of cross-species alignment while varying λ , for an extensile-contractile mixture. (b) Magnitude of phase separation, Δ , on varying λ for an extensile-contractile mixture.

4.3 Varying other parameters in the system

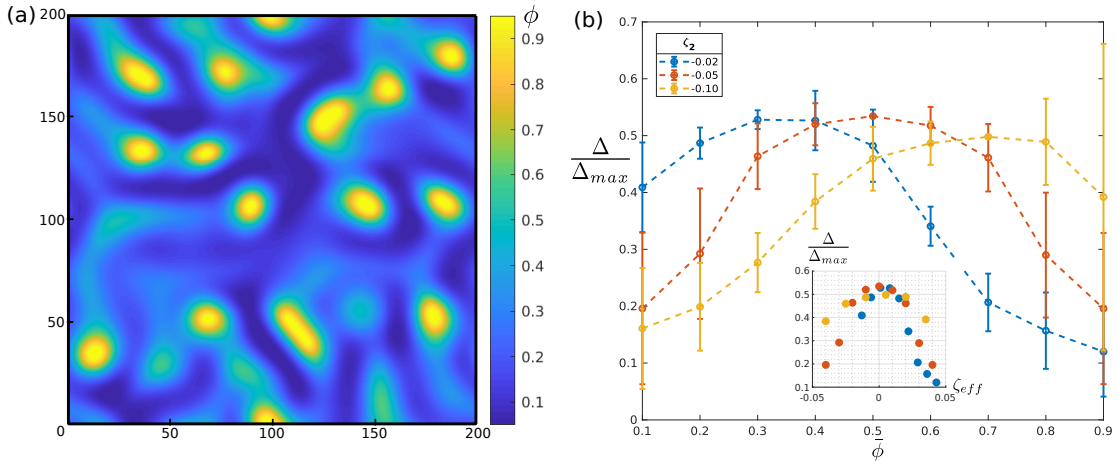


Figure 4.6: Changing Concentration Fraction: (a) Concentration field for a 30-70 mixture which phase separates into circular droplets ($\zeta_1 = 0.02, \zeta_2 = -0.005$, and $\bar{\phi} = 0.3$). The colourbar shows local concentration ϕ . (b) Normalised magnitude of phase separation, Δ/Δ_{max} , as a function of concentration fraction, $\bar{\phi}$, for three different activities ζ_2 , while holding $\zeta_1 = 0.05$ fixed. (Inset) Δ/Δ_{max} plotted as a function of effective activity $\zeta_{eff} = \zeta_1\bar{\phi} + \zeta_2(1 - \bar{\phi})$ is highest at low effective activity.

4.3.1 Concentration Fractions

We consider the effect of changing $\bar{\phi}$, the average of ϕ (concentration fraction of species 1), to study how the phase separation is affected if the system is not a 50-50 mixture of each species. On reducing $\bar{\phi}$, the elongated droplet network of Fig. 4.4(a) is replaced by small droplets of fluid 1 in a background of fluid 2 as shown in Fig. 4.6(a). At lower activities or higher surface tensions, the droplets are rounder and the flows are less turbulent.

For a given pair of activities ζ_1 and ζ_2 , different concentration fractions $\bar{\phi}$ give different degrees of phase separation Δ . The maximum possible Δ for a perfectly phase-separated system with infinitely sharp interfaces is $\Delta_{max} = \sqrt{\bar{\phi}(1 - \bar{\phi})}$. Since this is dependent on $\bar{\phi}$, we quantify the magnitude of phase separation at different $\bar{\phi}$ using the normalized Δ/Δ_{max} .

A plot of Δ/Δ_{max} for three different values of ζ_2 is shown in Fig. 4.6(b) ($\zeta_1 = 0.05$). We note that stronger phase separation is achieved when the effective activity $\zeta_{eff} = \zeta_1\bar{\phi} + \zeta_2(1 - \bar{\phi})$ is small (See inset, Fig. 4.6(b)) as this suppresses the active turbulence which leads to droplets breaking up. This implies that the highest phase separation is observed when the more active component has a smaller concentration fraction.

4.3.2 Friction

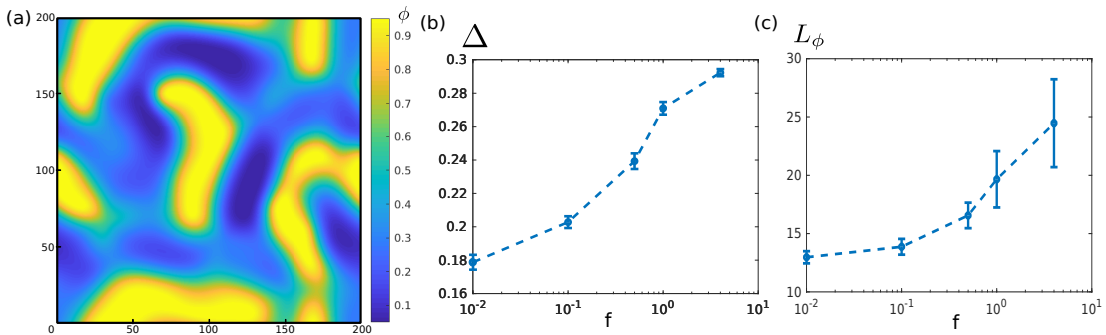


Figure 4.7: Changing Substrate Friction: (a) Concentration field for high substrate friction ($\zeta_1 = 0.05, \zeta_2 = -0.05, f = 4$), showing large, well-separated domains. The colourbar shows local concentration ϕ . (b) Magnitude of phase ordering, Δ , increases with higher substrate friction. (c) Size of phase-separated regions, L_ϕ , increases significantly with higher substrate friction.

Finally, we look at the effect of adding substrate friction f . On increasing friction, the active flows are weaker and less turbulent. The elongated droplet networks tend to aggregate to form more strongly phase-separated regions that are larger in size. A snapshot of the concentration profile is shown in Fig. 4.7(a). Fig. 4.7(b) shows the increase in the magnitude of phase separation upon increasing the substrate friction. The typical lengthscales of the phase-separated domains, L_ϕ , also increase with higher substrate friction (Fig. 4.7(c)).

At first glance, it might appear surprising that high substrate friction facilitates phase separation driven by active flows. However, although the overall dynamics are slower, the active turbulent flows are smaller in magnitude, and dissociation by active instabilities is very rare. Moreover, adding friction results in hydrodynamic screening of the active forces [27], reducing the correlation of the velocity at a point with forces applied on the fluid far away. In the extreme case of very high friction, the force balance condition changes and the velocity is now directly proportional to the applied force $\mathbf{u}^i \approx \mathbf{F}^i/f$ instead of $\nabla^2 \mathbf{u}^i \approx \mathbf{F}^i/\eta$. As a result, although the velocity field is weaker, velocities are better aligned normal to the interface, in the direction of the local active forces, as shown in Fig. 4.8.

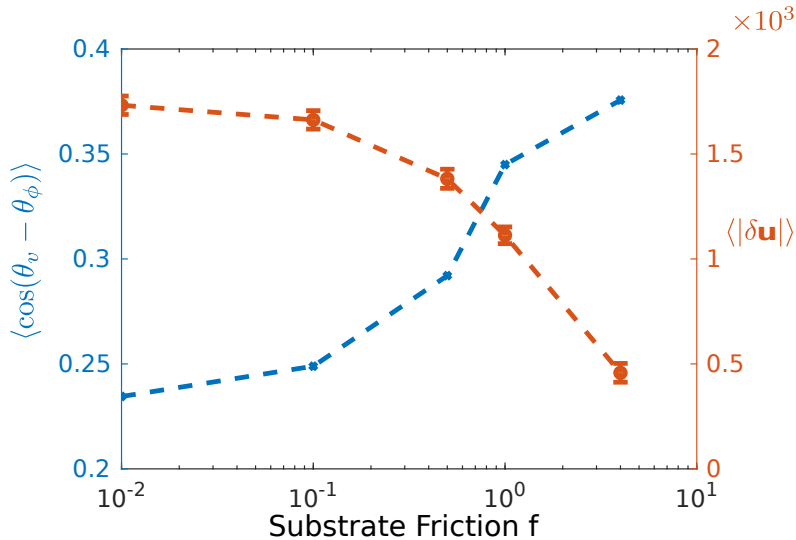


Figure 4.8: Variation of relative flow alignment and magnitude with increasing substrate friction. Left axis (blue): Relative flow between the fluid components orients strongly in the direction of increasing concentration gradient for higher substrate frictions. Right axis (orange): The magnitude of relative flow $|\delta \mathbf{u}|$ between the fluid components decreases with increasing substrate friction.

4.4 Summary

To summarise, we have extended a two-fluid model to study mixtures where both species are active nematics. We argued that even in the absence of any externally imposed elastic ordering between components, active nematics coupled by viscous drag tend to align parallel or perpendicular depending on the relative signs of the activities and the value of the flow-aligning parameter.

We observed turbulent microphase separation in mixtures of two active nematic fluids, each with a different activity. Imposing an elastic director alignment between the two active species plays a major role in determining the magnitude of the phase separation. In the absence of imposed parallel alignment, there is weak segregation which is most pronounced for a mixture of a highly active and a passive component. However, in the presence of imposed parallel alignment, coexisting domains comprised almost entirely of one component can be achieved with an extensile–contractile fluid mixture.

The magnitude and morphology of phase separation are also affected by other parameters in the system. As expected, varying the concentration fraction can change the morphology from elongated droplet networks to isolated circular drops. Perhaps more surprisingly, increasing substrate friction leads to stronger phase separation and a considerable increase in the size of the phase-separated domains.

Take-home message:

Mixtures of species with different intrinsic flows show the strongest phase separation.

“Ordo ab Chao”

(Translation: Out of chaos comes order.)

5

Active phase separation under confinement

In the previous two chapters, we have looked at phase separation in active mixtures undergoing active turbulence. In those systems, phase-separated regions form, move and disintegrate chaotically, driven by turbulent flows in the system. However, phase ordering in biology is often deterministic - with different cell types sorting themselves into specific positions. For instance, in starfish embryos, endodermal and ectodermal cells mixed together will spontaneously sort themselves, with the endodermal cells migrating inwards and the ectodermal cells migrating to the outer edge of the embryo [75] (Fig. 3.1a–c). In studies of cancer tumours, a co-culture of metastatic breast cancer (MDA-MB-231) and non-tumorigenic epithelial (MCF-10A) cells shows architecture inversion, with the metastatic cells surrounding the epithelial cells two days after seeding but the non-tumorigenic cells switching positions to surround the metastatic cells two days later (four days after seeding) [105].

Thus, it is important to study phase separation in active systems confined by boundaries. While active nematics in bulk are always unstable to active turbulence [34], confined active nematics can show ordered flow states, notably spontaneous laminar flows inside channels [30], and spontaneous rotational flows in droplets [35].

Therefore, we investigate active phase separation in mixtures with ordered flow states, and in active turbulent mixtures in confinement. The rest of the chapter is divided as follows: In Sec. 5.1, we show that turbulent active nematics preferentially

aggregate or de-aggregate from the walls depending on the anchoring of the nematic field at each wall. In Sec. 5.2, we show that the active component migrates towards the centre of the channel in active channels with laminar flows. Finally, in Sec. 5.3, we show that the more active material accumulates at the outer boundary for droplets in viscous confinement. In each of these cases, we investigate the mechanism behind phase ordering and demonstrate how gradients in the magnitude of nematic order, nematic orientation field and even a preference towards active anchoring can lead to coherent sorting in confined active nematics.

5.1 Anchoring at Walls Controls Active Phase Separation

Consider a homogeneous mixture of an active nematic and a passive nematic fluid confined in a box, with free slip at the boundaries, and homeotropic or planar anchoring specified on the box walls. The orientation field is fixed (anchored) at the boundary by specifying Dirichlet conditions on the $\mathbf{Q}$ field, and is a form of strong anchoring - we shall call this ‘thermodynamic anchoring’. The magnitude of activity is high enough that the active nematic fluid inside the box is in a turbulent state.

We observe that the concentration of the active nematic at the boundary increases if the thermodynamic anchoring has the same orientation as active anchoring. Thus, extensile (contractile) nematics have a higher concentration at the wall than in bulk if the thermodynamic anchoring is planar (homeotropic). On the other hand, if the thermodynamic anchoring has the opposite orientation as active anchoring, then the active material has a lower concentration at the wall. Thus, extensile (contractile) nematics have a lower concentration at the wall if the anchoring is homeotropic (planar). This is shown in Fig. 5.1.

Moreover, the concentration of active material at the wall increases linearly with activity when the thermodynamic anchoring matches active anchoring. For opposite anchoring, however, the active concentration at the wall depletes until saturating at a certain concentration upon increasing activity.

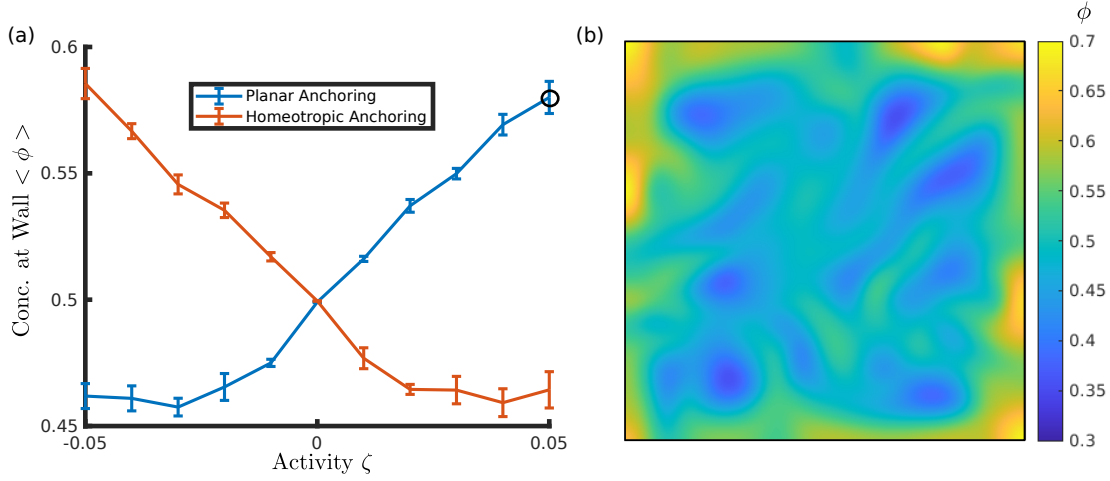


Figure 5.1: (a) Concentration fraction of active material at the walls, for a mixture of an active nematic and a passive nematic species undergoing active turbulence. The concentration of active material at a wall is enhanced for extensile nematics with thermodynamic planar anchoring, and for contractile nematics with homeotropic anchoring. The active material is depleted from the walls when the thermodynamic anchoring is reversed. (b) Snapshot of the concentration field for $\zeta = 0.05$, and planar anchoring (marked by a black circle in panel (a)). Colourbar shows the concentration of the active species, showing preferential accumulation near the boundaries.

Analogously to Chapter 4, we can study the same effect when we have a mixture of two active species. If both species have the same sign of activity, the more active species shows the same concentration enhancement or depletion trend, with the magnitude of the effect depending on the difference in activities. Due to overall incompressibility, the less active species shows the opposite trend.

This effect is greatly enhanced when we mix extensile and contractile active nematics with imposed elastic alignment. In this case, the extensile (contractile) species accumulates at the walls under planar (homeotropic) confinement, and the other species simultaneously migrates away from the wall. These effects reinforce each other, magnifying the magnitude of phase ordering at the wall (Fig. 5.2).

Mechanism

Analogous to Sec. 3.3.1, phase separation is driven by an imbalance of active forces at an active gradient. Since the active stress is given by $\zeta\phi Q$, these active forces can be due to gradients in concentration, nematic order and activity. Moreover,

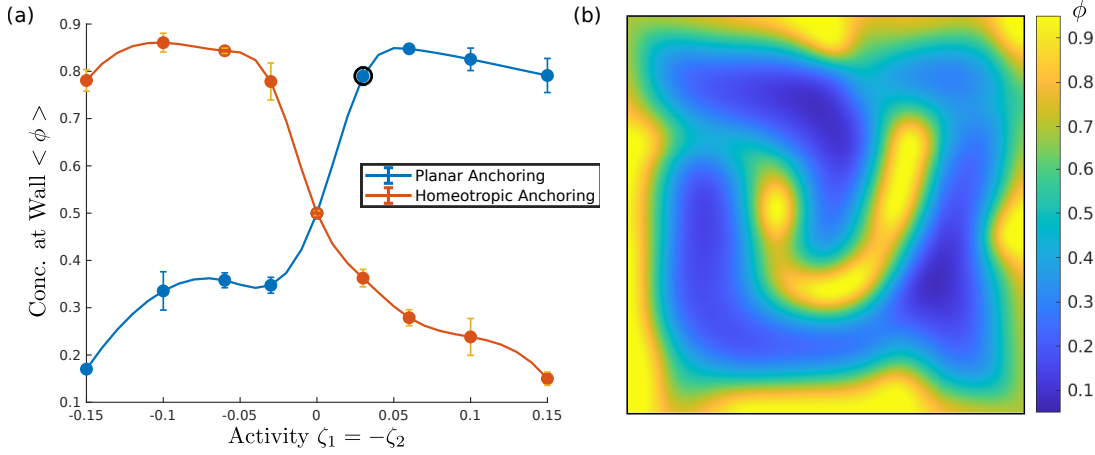


Figure 5.2: (a) Concentration fraction of active extensile material at the walls, for an extensile-contractile mixture with equal activity. The degree of active phase separation at the wall is significantly higher than in the active-passive case. (b) Snapshot of the concentration profile for an extensile-contractile system with $\zeta_1 = 0.03, \zeta_2 = -0.03$ (marked by a black circle in panel (a)), with planar anchoring at the walls. Colourbar shows the concentration of the extensile fluid, which accumulates preferentially at the boundaries of the box. The bulk also shows active phase separation, as described in Chapter 4 .

the direction of these forces is dictated by the direction of the orientation field and a concentration gradient.

We study a box, where the thermodynamic anchoring at the walls aligns the nematics at the angle θ_{anchor} relative to the wall. The director field at the boundary is aligned and ordered, while in the bulk it is turbulent and chaotic. First, let us consider the case where the concentration of active material is higher at the wall. The active force along the normal to the wall is given by $\zeta \nabla(\phi S) \cos(2\theta_{\text{anchor}})$ in the direction of the wall, where we assume that the gradient of ϕS is normal to the wall. If $\zeta > 0$ (extensile) and $\theta_{\text{anchor}} = 0^\circ$ (planar), this results in an active force towards the wall, which drives active flows increasing the concentration of active material at the wall. This increases the concentration gradient and makes active forces stronger. This process continues until the accumulation is stopped by the restoring free energy, and by chaotic active turbulent flows mixing up the regions. The mechanism is analogous for (contractile) $\zeta < 0$, with $\theta_{\text{anchor}} = 90^\circ$ (homeotropic). In the case of $\zeta > 0, \theta_{\text{anchor}} = 90^\circ$ or $\zeta < 0, \theta_{\text{anchor}} = 0^\circ$, the active flows point away from the wall instead, leading to a reduction in the concentration of the active material at

the boundaries. This process continues until the lower active concentration at the boundary compensates for the higher nematic order S at the boundary, leading to the depletion of active concentration saturating at higher activities.

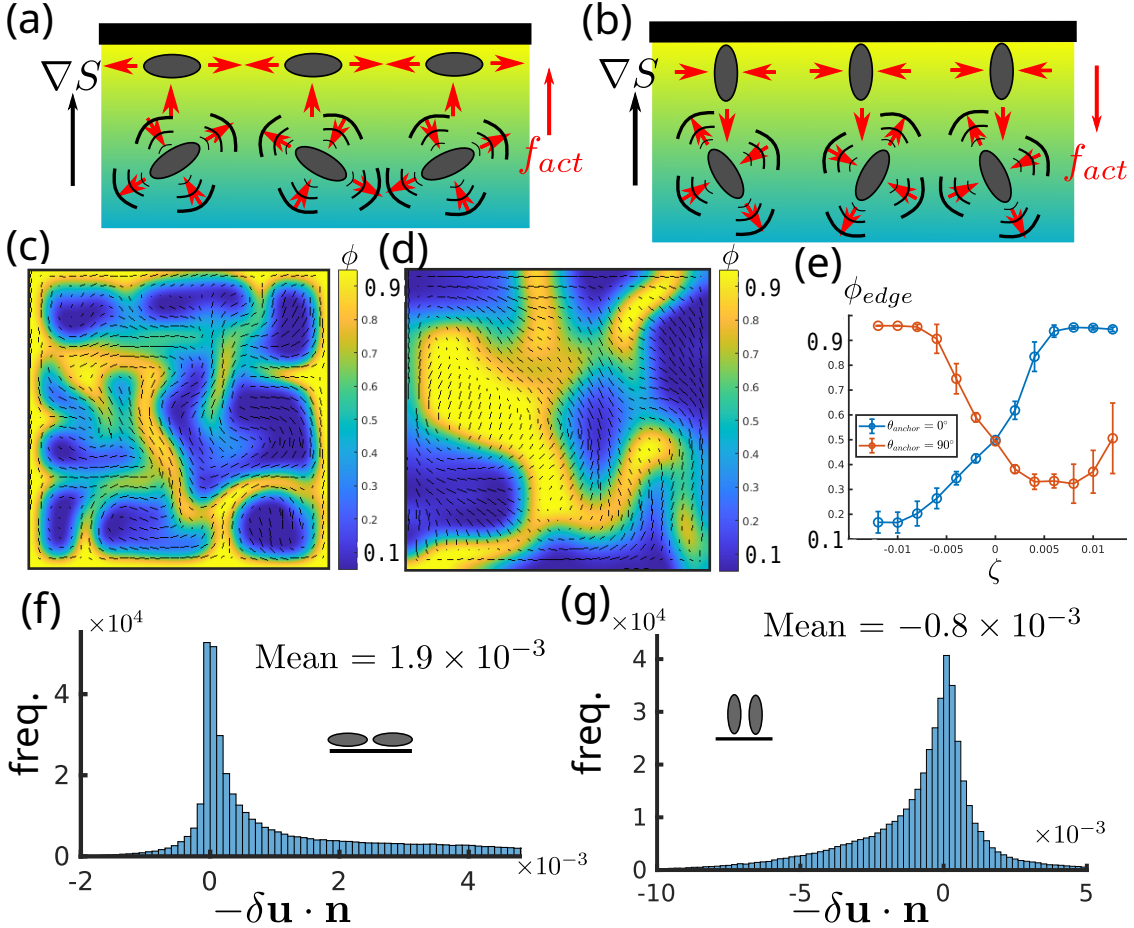


Figure 5.3: (a)-(b) Schematic showing nematic order gradient and active forces at an anchored boundary. Extensile nematics are ordered at the boundary, but orient chaotically in the bulk (shown by curved black lines), resulting in a gradient in S . The resultant active force f_{act} points (a) towards the boundary for planar anchoring, and (b) away from the boundary for homeotropic anchoring. (c)-(e) An active nematic confined inside a box, with free slip and planar anchoring at the walls. (c) For extensile activity with planar anchoring, the concentration of active nematic at the boundary is enhanced. (d) For an equal magnitude of contractile activity with planar anchoring, the concentration of active nematic at the boundary is depleted. (e) Active nematic concentration is much higher for larger order gradients at the wall. The asymmetry between extensile and contractile is due to the non-zero flow tumbling parameter. (f)-(g) Distribution functions of the relative active flow towards the walls, $-\delta \mathbf{u} \cdot \mathbf{n}$, for an extensile nematic mixture confined in a square box. (f) For planar anchoring, there is a net flow of active material towards the wall. (g) For homeotropic anchoring, there is a mean flow of active material away from the wall.

We still need to address how the difference in concentration at the boundary is set up in the first place. Initially, the flow is turbulent and chaotic everywhere and the orientation field fluctuates rapidly, leading to no net active force averaged over time. However, the orientation field near the boundary has a fixed direction due to thermodynamic anchoring, leading to a net active force pointing towards (away) from the boundary if the thermodynamic anchoring has the same (opposite) sign as active anchoring. This active force is coherent and does not vanish on averaging over time. This active force can lead to a net migration of the active fluid, leading to a concentration gradient at the walls of the container. This mechanism for using an order gradient to initially set up a concentration gradient is shown in Figs. 5.3a–b.

One consequence of this mechanism is that the degree of wetting is stronger in systems with higher order parameter gradients. We verify this by using the parameters $C^1 = 0.002$, $K^1 = 0.02$, $\lambda = 0.7$ which has a longer passive nematic lengthscale [63]. In Figs. 5.3c–e, we show the qualitative difference in concentration field upon only changing the sign of the activity. We also verify the mechanism numerically by measuring the relative flow towards to the box walls $-\delta\mathbf{u} \cdot \mathbf{n}$ (where $\mathbf{n}$ is the normal vector of the wall pointing inwards). We find that extensile nematics have a relative flow towards the walls when the anchoring is planar (Fig. 5.3f), and away from the walls when the anchoring is homeotropic (Fig. 5.3g).

5.2 Laminar channel flows

Active nematics inside channels are a paradigmatic example of confined active matter, with unidirectional and bidirectional laminar flow [30], oscillatory flow, vorticity lattices [106] and active turbulence [107]. We consider a homogeneous mixture of an active nematic and a passive isotropic fluid inside a channel with parallel director anchoring and free slip at the channel walls. We study the evolution of the concentration field for the bidirectional flow state just above the linear flow transition [30].

In this configuration, the active material migrates away from the walls and towards the centre of the channel. The stripe in the centre of the channel is highly

active and can further undergo active instabilities, transitioning into an oscillatory flow or vorticity lattice structure, with the associated characteristic velocity profiles in the combined fluid. The concentration fields and velocity profiles observed in a channel on increasing activity are shown in Figs. 5.4a–c.

Fig. 5.4a shows the concentration field migrating to the centre of the channel in the bidirectional laminar flow state. The degree of phase separation depends both on the activity and on the width of the channel. At higher activities, the flow in the channel transitions from laminar flow to a dancing state with vorticity lattices, shown in Fig. 5.4b.

At even higher activities, the flow in the channel transitions to active turbulence. In this state, the active material chaotically aggregates in the bulk (Fig. 5.4c), due to the mechanism described in Sec. 5.1.

Mechanism

Let us consider a channel of width L in the y direction ($y \in [0, L]$), and homogeneous in the x -direction. The channel contains a mixture of *contractile* nematic ($\zeta < 0$) and passive isotropic fluid. Flows in the channel are along the x direction.

For a single fluid, the bidirectional laminar flow configuration has an orientation field given by

$$\theta(y) = \epsilon \sin(ky) , \quad (5.1)$$

where $k = \pi/L$ is the wavenumber of the first unstable mode, and ϵ the amplitude of the perturbation. This is also the first unstable mode for a uniformly mixed fluid mixture initialized at rest, since the relative flow driving phase separation is much slower than flows in the centre of mass fluid.

This orientation field generates active forces along the channel driving the bidirectional flow, however, there is also a component across the width of the channel. This component is given by

$$F_y = -\zeta \partial_y (\phi Q_{yy}) = -\zeta S \left[-(\partial_y \phi) \cos(2\theta) - \phi \partial_y (\cos 2\theta) \right] , \quad (5.2)$$

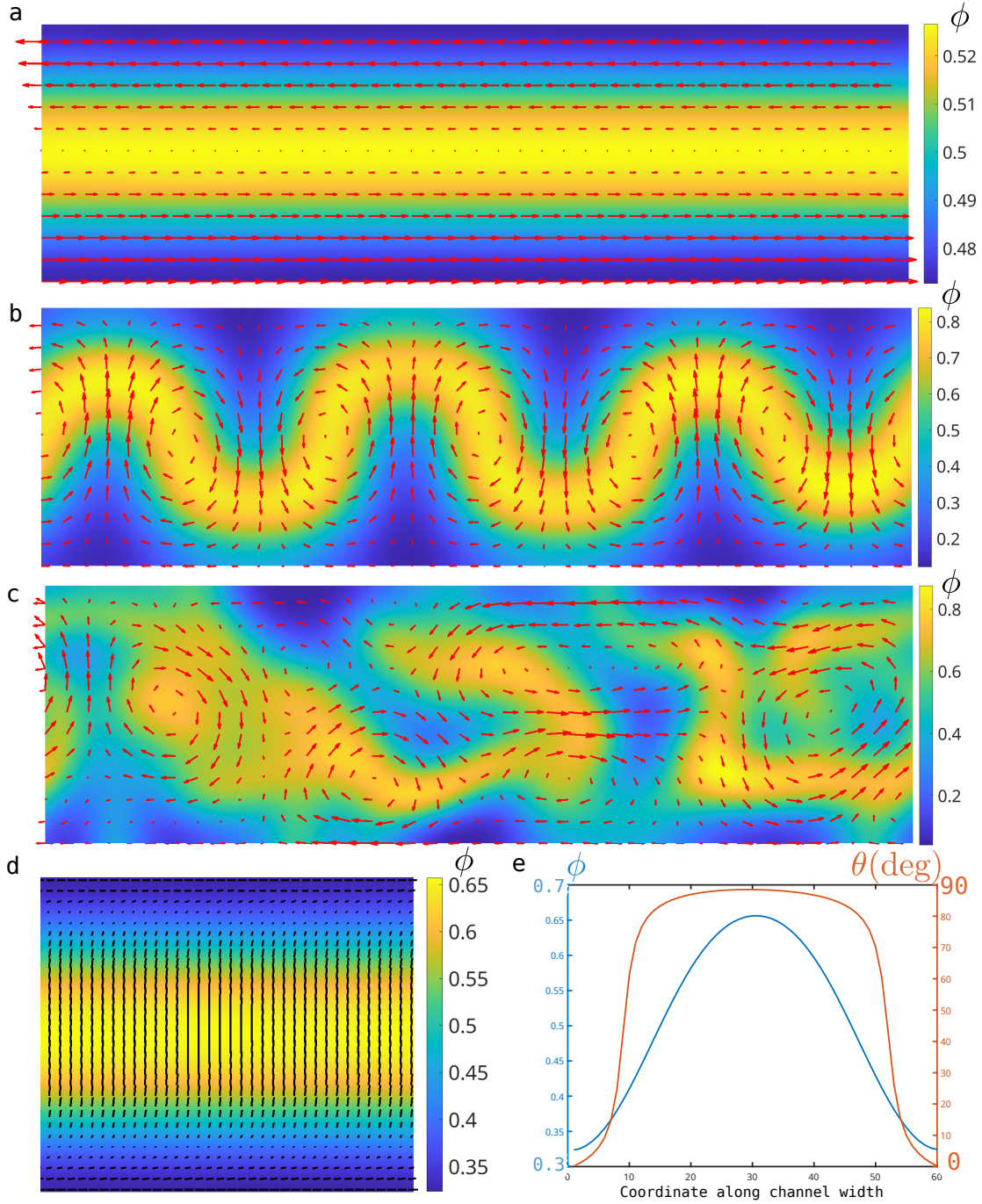


Figure 5.4: Active phase separation in a contractile and passive fluid mixture in a channel, with planar anchoring at the wall. (a)–(c) Concentration field (ϕ) and velocity field ($\mathbf{u}$) obtained on increasing activity. The colourbar shows ϕ , and the red arrows show $\mathbf{u}$. (a) A contractile band in the middle of the channel forms at low activities close to the spontaneous flow transition. (b) At higher activities, the channel enters the dancing state [106], and the concentration field forms a bend structure. (c) At higher activity, the flow enters an active turbulent state, with higher active concentration in the middle of the channel. (d)–(e) shows the orientation field and concentration field in the band-forming regime at low activities. In panel (d), black lines denote the director field $\mathbf{Q}$, and the colourbar shows the concentration ϕ .

where we have assumed that the magnitude of nematic order S is held constant throughout the channel. When the fluids are homogeneously mixed, the first term, proportional to $\partial_y \phi$, is zero, and the active force in the y direction is $-2\zeta\phi Sk\epsilon \cos(ky) \sin(2\theta)$. Since $\zeta < 0$, this force in the active component fluid drives material towards the centre of the channel. Once a concentration gradient has been set up, however, the first term in Eq.(5.2) yields $\zeta S \cos(2\theta)(\partial_y \phi)$, setting up active forces which oppose the concentration gradient.

From Eqs.(5.1) – (5.2), the net active force across the channel for a bidirectional laminar flow is given by

$$F_y(y) = -2\zeta S [\phi k \epsilon \cos(ky) \sin(2\theta) - \cos(2\theta)(\partial_y \phi)]. \quad (5.3)$$

The magnitude of the force is dependent on the angle of the director field θ . We plot the director field overlaid over the concentration field ϕ in Figs. 5.4e–f.

The degree of phase separation also depends on the width of the channel, with higher phase separation for wider channels with laminar flow. At higher activities, the coherent director and concentration profiles give way to active turbulence. The concentration of contractile active material is depleted at the walls, where the director is anchored planar to the walls. The mechanism for this behaviour is discussed in Sec. 5.1.

5.3 Droplet in viscous confinement

Finally, we look at active droplets confined inside a highly viscous surrounding fluid, in a regime where the first component is isotropic in the absence of activity ($S = 0$). We impose continuity of stress boundary conditions across the interface, in addition to no-penetration conditions and continuity of the tangential component of the velocity field across the interface [108]. Inside the droplet, we start with a homogeneous mixture of an active nematic fluid mixed with a passive isotropic fluid.

In order to simulate the boundaries in this setup, we use an additional phase-field φ , initialized as a circle, to demarcate the active mixture inside ($\varphi = 1$) from the viscous fluid outside ($\varphi = 0$). The boundary was then allowed to relax to

a free-energy minimum (Eq.(2.22) with $A_\varphi = 0.4, K_\varphi = 0.1$) to create a smooth interface, and then the field φ was kept fixed for the simulation. The interior of the droplet was initialized with uncorrelated patches of local nematic order, with the orientation chosen randomly and uniformly between the angles 0 and π .

Past research [35] has shown that for a range of activities, active nematic droplets confined in this manner spontaneously rotate. The magnitude of nematic order is high near the edge, and zero in the middle. There is a +1 topological defect in the middle of the droplet, which is ‘virtual’ since it forms in a region without nematic order [35].

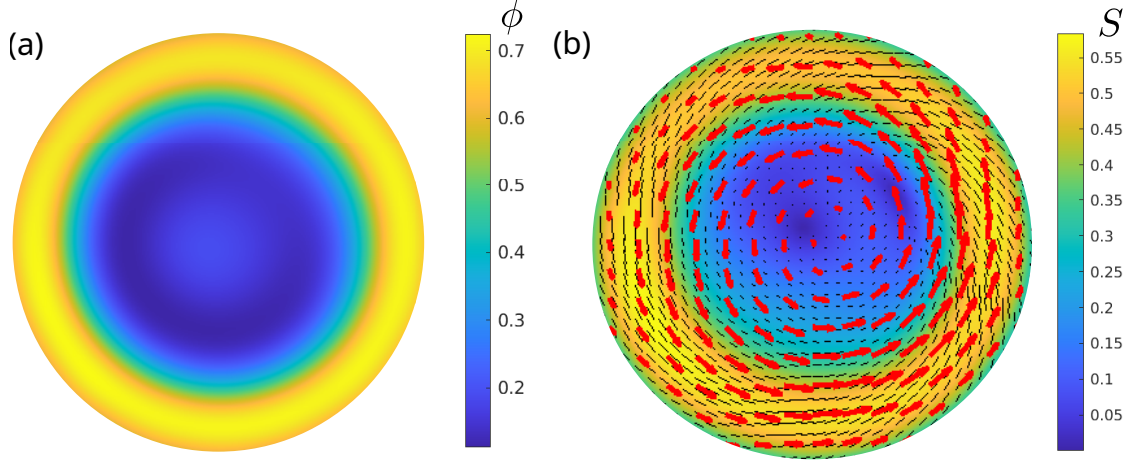


Figure 5.5: Active phase separation in a spontaneously rotating droplet with an initially uniform mixture of active and passive fluid ($\zeta_1 = 0.005, \zeta_2 = 0$). (a) The active fluid preferentially accumulates at the droplet boundary. The colourbar denotes the concentration of active fluid. (b) The nematic director field of the spontaneously rotating droplet is shown in black, while red lines show the velocity field in the combined fluid. Colourbar denotes the magnitude of nematic order in the active fluid.

In a mixture of an active nematic and a passive fluid, we observe that the spontaneously rotating state develops with the active material preferentially accumulating at the boundary, forming a ring of active fluid around the passive fluid. This is shown in Fig. 5.5.

At much higher activities, droplets go into an active turbulent state. In this case, phase separation at the boundary is much lower and less uniform, as turbulent flows mix up the resultant phases. Due to active anchoring at the droplet edge, however, active fluid still preferentially accumulates at the boundaries. This is

driven by mechanisms similar to Sec. 5.1.

Mechanism

An active droplet confined by a viscous environment develops nematic order at the confining boundary[109–111]. If the active material is isotropic in the bulk, this sets up a gradient of nematic order, ∇S , oriented towards the boundary of the active droplet.

Due to the orientation of the nematic field at the boundary, the gradients in S generate flows in the active fluid towards the droplet edge. This increases the concentration of active material ϕ at the boundary, setting up a concentration gradient which further amplifies the active force in the direction of the boundary. This sets up an instability, leading to the formation of an active ring at the edge of the droplet, with the passive material in the centre.

5.4 Summary and Discussion

In this chapter, we showed how mixtures of active nematic and passive isotropic species under confinement can spontaneously phase order, sorting the active component into specific regions. In a channel with laminar flows, the active component migrates towards the centre of the channel. In a droplet with spontaneous rotation, however, the active component sorts to the outer boundary. In active turbulent states under confinement, the active material preferentially accumulates towards or away from the boundary depending on the nematic anchoring at the walls. Our results are relevant for studying confined active nematic systems e.g. a confined mixture of microtubules and motors proteins in a solvent [112]. Our results also highlight a potential mechanism to sort active nematic cell types towards or away from a boundary using active forces from cell motility or growth.

In Chapters 3–5, we have discussed a new mechanism of phase separation driven by active nematic flows. Our results add to a growing body of work describing the possibility of phase separation driven by activity, here focusing on active flows in fluid

mixtures. Cell ordering and sorting in embryogenesis, and compartmentalisation into different cell types, are important biological processes where activity may play a role [82, 103, 104, 113, 114]. Our work may also be relevant in understanding organization and movement in multi-species bacterial colonies [115, 116], or the intracellular liquid-liquid phase separation [72, 117] which results in membrane-less organelles.

Take-home message:

In confined active nematic mixtures, phase ordering sorts the components into patterns.

“I believe that at the end of the century the use of words and general educated opinion will have altered so much that one will be able to speak of machines thinking without expecting to be contradicted.”

— Alan Turing (1950)

6

Coupling Turing stripes to active flows

We now change topics and look at the interplay of chemical signalling with an active nematic material. We first introduce Turing patterns, the archetype for chemical patterning, and couple them to active flows through advection by the nematic background. We find that active flows can affect the morphology and strength of Turing stripes, bending and breaking stripe segments until the patterns ultimately dissolve at high activities. We quantify the balance of patterning and active flows by a single Karlovitz number, characterizing the relative timescales of active flows and the reaction flux. We then show that when the Turing stripes with different morphogens have extensile and contractile activity respectively, the system self-organizes into a configuration with shearing flows inside each stripe. Characteristic active instabilities make the stripes unstable to fracturing and slipping sideways to join their neighbours. Our results are an example of the non-trivial role of mechano-chemical coupling in active systems.

6.1 Introduction

Chemical signalling is one of the key drivers of patterning, and plays a crucial role in tissue morphogenesis. Following Turing’s seminal paper [36] showing that coupled reaction-diffusion equations can lead to pattern formation, there have

been many interesting results extending his ideas to more closely model biological processes [118–123]. The effect of bio-chemo-mechanical feedback on active shells has been studied recently [124], as has the coupling of a biphasic model of tissue mechanics with morphogen reaction and diffusion [125]. The robustness of Turing-pattern formation in biological systems under the effects of noise and feedback is also an active area of research [126].

It is now timely to investigate the interplay between flows arising from active stresses and the pattern-formation driven by chemical reaction-diffusion processes. Siero *et al.* have discussed how Turing patterns respond to advective flows [127]. There have also been studies on coupling pattern formation to active flows in one dimension [128], where the existence of a region of non-trivial pattern formation as an interplay of activity and reaction-diffusion was discovered. This work was extended to show the existence of pulsatory patterns when an activator-inhibitor system of two chemicals was used in higher dimensions [129]. Recent work has shown that coupling mechanical activity driven by chemical signals with geometry and flows can give rise to non-trivial shapes and dynamics on active surfaces [130].

In this chapter, we study the coupling between Turing patterns and active turbulence, the chaotic flow state characteristic of active nematics [27]. Working in two dimensions, we consider a two-species, reaction-diffusion model which orders into Turing patterns, where each of the chemical species can induce local active stresses. The equations describing two different classes of Turing patterns and how to solve them numerically are described in Sec. 6.2. In Sec. 6.3, we discuss how activity can affect the Turing stripe patterns formed by the reaction-diffusion system. We find that stripes bend, break up into patches and then completely disappear on progressively increasing the activity. Then, in Sec. 6.4, we study a specific coupling, where the two chemical species induce activities which are equal in magnitude, but take different signs. This leads to a state with alternating shear flows across Turing stripes, which undergo slipping at longer timescales. In Sec. 6.5, we discuss the solution for more general activity assignments, highlighting the role of active instabilities.

6.2 Turing Patterns

We consider an active nematic system with flow field $\mathbf{u}$ and orientation field $\mathbf{Q}$, coupled through the usual equations of active nematic hydrodynamics (Eqs.(2.2), (2.1), (2.12), and (2.20)). In addition to the active nematic, we add scalar fields c_A and c_B describing the concentrations of two morphogens, A and B, which reside within the active fluid. Their dynamics are modelled by

$$\partial_t c_A + \nabla \cdot (c_A \mathbf{u}) = D_A \nabla^2 c_A + R_A(c_A, c_B), \quad (6.1)$$

$$\partial_t c_B + \nabla \cdot (c_B \mathbf{u}) = D_B \nabla^2 c_B + R_B(c_A, c_B) \quad (6.2)$$

Here, the term $\nabla \cdot (c_{A,B} \mathbf{u})$ describes the advection of each morphogen by the flow field $\mathbf{u}$ of the active nematic. The right hand sides models the reaction and diffusion dynamics of the morphogens. D_A and D_B are diffusion constants, while $R_A(c_A, c_B)$ and $R_B(c_A, c_B)$ are functions modelling the creation and interaction between the morphogens.

We have modelled our system with a single fluid field with two additional concentration fields representing the morphogens. It would be interesting to revisit this chapter with the two-fluid model described in the previous sections, adding reaction terms between the two components fluids to model Turing patterns.

6.2.1 Schnakenberg model

We use the Schnakenberg model [36, 131, 132], which is a minimal model to generate Turing patterns. In this case, we choose

$$R_A(c_A, c_B) = \gamma_c(\alpha_C - c_A + c_A^2 c_B), \quad (6.3)$$

$$R_B(c_A, c_B) = \gamma_c(\beta_C - c_A^2 c_B) \quad (6.4)$$

where α_C , β_C and γ_c are model parameters. In the absence of flow the phase space of Eqs. (6.1–6.2) consist only of homogeneous, stripe-forming (see Fig. 6.1a), and spot-forming regions [133], with the non-trivial Turing patterns appearing for imbalanced diffusion constants, $d_C \equiv D_B/D_A \gtrsim 10$ or $\lesssim 0.1$. The concentration

profiles of the two species are out-of-phase. The characteristic lengthscale of the Turing spots or stripes follows from linear stability analysis as [134]

$$L_{Turing} = 2\pi \sqrt{\frac{D_A}{\gamma_c}} \left[\frac{(\alpha_C - \beta_C)/(\alpha_C + \beta_C) + (\alpha_C + \beta_C)^2}{d_C - 1} + \frac{d_C + 1}{d_C - 1} \sqrt{\frac{2\beta_C(\alpha_C + \beta_C)}{d_C}} \right]^{-\frac{1}{2}}. \quad (6.5)$$

In the simulations, we will change the rate of reaction γ_c to control the contrast and width of the Turing patterns. For small γ_c , the rate of reaction is much lower than the rate of diffusion, and we find wide stripes with a small difference in morphogen concentration between consecutive stripes. For larger values of γ_c , a strong reaction rate leads to thinner stripes with a higher variation in morphogen concentration.

6.2.2 Gierer-Meinhardt model

We can also use reaction-diffusion systems to generate in-phase Turing patterns. For this, we use the Gierer-Meinhardt model [120, 123], choosing the reaction terms

$$R_A(c_A, c_B) = \gamma_c \left(\left(X_0 + \frac{c_A^2}{1 + K_A c_A^2} \right) / c_B - \mu_A c_A \right), \quad (6.6)$$

$$R_B(c_A, c_B) = \gamma_c \left(c_A^2 - \mu_B c_B \right). \quad (6.7)$$

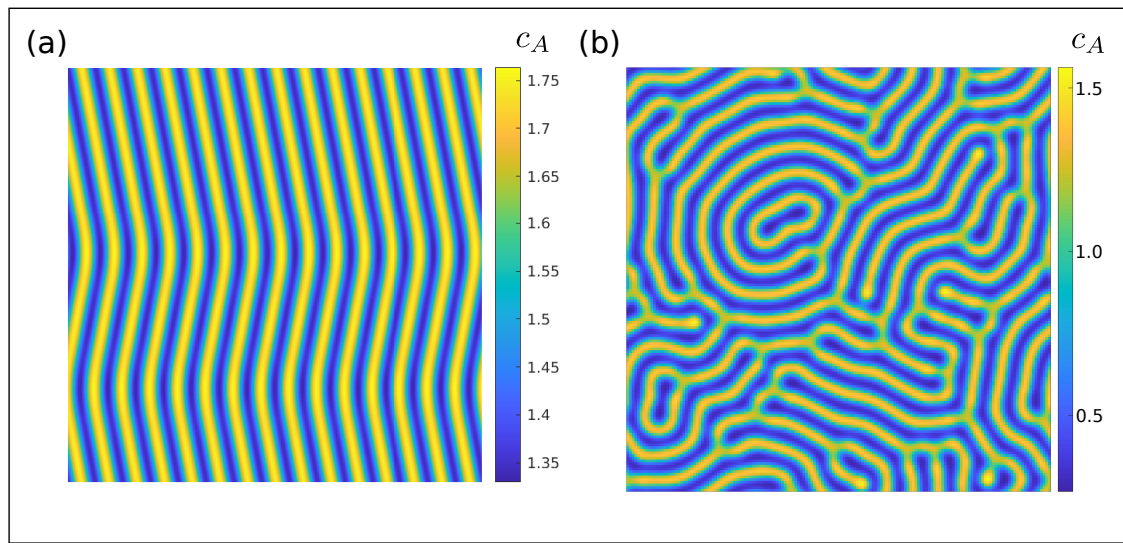


Figure 6.1: (a) Turing stripes obtained from the Schnakenberg model. (b) Turing stripes obtained from the Gierer-Meinhardt model. Colourbar shows the concentration field of c_A in both cases.

In contrast to the Schnakenberg case, the Gierer-Meinhardt stripes do not align globally, but have a fixed concentration profile where stripes with different orientations join together (Fig. 6.1b).

6.2.3 Simulation details

We solve the equations of motion using the hybrid lattice Boltzmann method (Appendix. A). In addition, we solve for the concentration field, Eqs. (6.1, 6.2), using a first-order finite difference scheme. These concentration solver was benchmarked by comparing analytical solutions for diffusion and reaction equations separately, and reproducing a wide variety of reaction-diffusion patterns [135, 136].

The simulations were run on a grid size of $L_x = 160$, $L_y = 160$ lattice units in the x and y directions respectively. Periodic boundary conditions were used in both x and y . Simulations were typically run from a uniform concentration profile with small added noise, or a random initial configuration. However, we checked that the same results hold for a wide variety of initial conditions of concentration and nematic director field.

Parameters used for the active nematic were $\rho = 40$, $\Gamma = 1$, $K = 0.15$, $A = -0.1$, $B = 0$, $C = 0.05$, $\lambda = 0.3$, $\eta = 1$, $P = \rho/3$ and $S_{nem} = 1$, and lattice Boltzmann time step, $\Delta t_{LB} = 1$, unless otherwise specified. The activity ζ was varied within the range $[0.01, 0.20]$.

For the reaction diffusion system, we used a Schnakenberg model (Eqs.(6.3), 6.4) with $D_A = 1.0$, $D_B = 20.0$, $\alpha_C = 0.14$, $\beta_C = 1.41$ corresponding to the stripe forming regime [133], and γ_c within the range $[0.1, 1.0]$ to vary the strength and contrast of the Turing stripes. The time step was $\Delta t_{LB}/200$. The shorter time step is needed to solve for the concentration profile sufficiently accurately in the regime where the pattern formation mechanism is roughly comparable in strength to the advection i.e. the Turing lengthscale is comparable to the lengthscale of the active nematic instability.

We also verified that our results hold for the Gierer-Meinhardt model (Eqs.(6.6), 6.7). For this system, we used the parameters $\mu_A = \mu_B = 5$, $\gamma_c = 0.01$, $D_A =$

0.2, $D_B = 10$, $X_0 = 0.1$, and $K_A = 0.25$ and $\Delta t = \Delta t_{LB}/200$ instead. This creates in-phase Turing patterns with parallel stripes spaced 20 lattice units apart, with a concentration difference $c_A \in [0.27, 1.56]$, and $c_B \in [0.14, 0.20]$.

6.3 Turing patterns advected by active flows

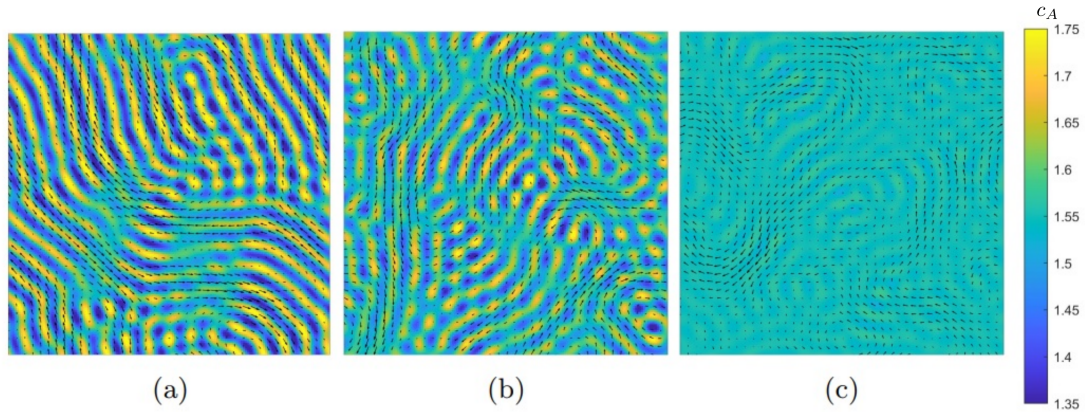


Figure 6.2: Increasing the strength of activity in a coupled Turing-active nematic system. The colour bar denotes the distribution of the c_A profile and the velocity field is depicted by arrows. (a) $\zeta = 0.04$, long bent stripes; (b) $\zeta = 0.07$, stripe segments; (c) $\zeta = 0.17$, no Turing stripes.

For the active reaction-diffusion system the choice of parameters listed above creates a Turing pattern of parallel stripes of width 5.5 lattice units each. The concentration field of the morphogens alternates between high and low values across adjacent stripes, with the concentration of the first morphogen c_A ranging from 1.35 to 1.75 with a mean value of 1.55, and the second morphogen $c_B \in [0.54, 0.64]$ with a mean value of 0.59. With our simulation parameters and in the absence of activity, the concentration fields show a clear sinusoidal variation profile across the different stripes (see Fig. 6.6b).

We first consider the case where the morphogens are advected by the flow field of the active nematic but the nematic is not affected by the presence of the Turing pattern. Stresses due to the director field of the active nematic create the typical flow and vortices associated with active turbulence [137]. These flows in turn advect the concentration field, and affect its properties. Fig. 6.2 compares examples of the different behaviours of the concentration fields as the activity ζ is varied.

At low values of activity, Turing stripes emerge and span the entire system. These bend along the direction of the active flow, but are not broken apart (Fig. 6.2a). This is possible because the vortices generated by the active flux are large in scale, the field lines bend slowly, and the velocities are small. On increasing the activity, the flow is able to overcome the ordering and the stripes break apart to form spots and stripe segments. These patches have a high variation in morphogen concentration (Fig. 6.2b) and are advected around by the nematic flow. On further increasing the activity, Turing stripes no longer form because the advective flux generated by the activity is sufficient to destroy any patterning generated by the reaction-diffusion flux, henceforth called the *Turing flux* (Fig. 6.2c). The same sequence of phases is seen if the reaction strength γ_c is decreased at fixed activity. Decreasing γ_c decreases the strength of Turing patterns, and consequently lower active flux is required to break them. We have verified that the transition point does not change on changing the elasticity or the flow aligning parameter of the system.

A Karlovitz number can be defined as the ratio of the timescale of the reaction equation (the *reaction timescale*) to an advective timescale [138]. The advection is driven by active forces, and the timescale of advection is given by $t_{adv} = l/U$, where l is a characteristic lengthscale for the active nematic and U is a characteristic velocity [128]. For an active nematic, $U \sim \zeta l/\eta$ [63]. Hence, $t_{adv} \sim \eta/\zeta$. When the system is close to forming Turing patterns, the Turing flux is mostly controlled by the reaction term, γ_c . Hence, the Turing timescale is proportional to $t_{Turing} \sim (\gamma_c)^{-1}$ and we expect the crossover between different regimes to be controlled by the Karlovitz number, $Ka \sim \zeta/(\eta\gamma_c)$.

To demonstrate this we note that, if the system exhibits Turing patterns, local increases in concentration must be much larger than the fluctuations in concentration. Therefore the patterns can be characterized by measuring the fraction of the system where the concentration of morphogen A, c_A , is greater than a given cutoff value which we take to be 1.68. The maximum concentration is 65% of the Turing value in the absence of advection for this cutoff, however we have checked that the conclusions are independent of the value chosen. Further details can be found in

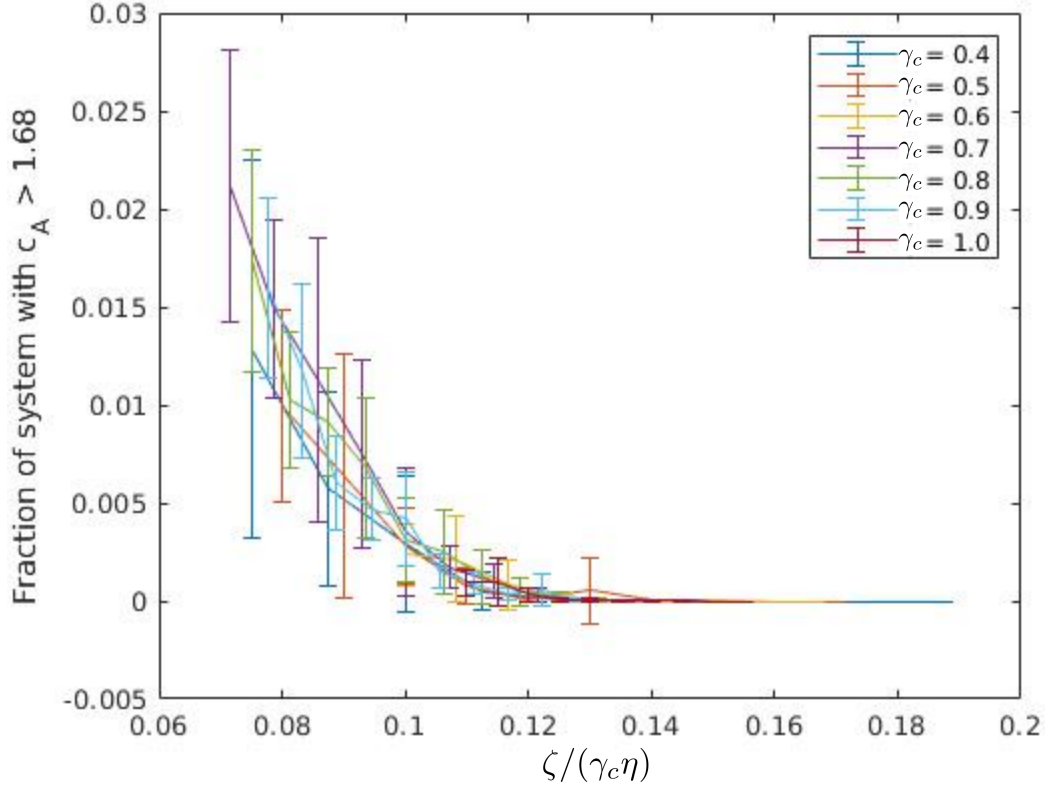


Figure 6.3: Fraction of active Turing system with concentration of A species above a chosen threshold as a function of Karlovitz number for different values of the Turing reaction strength, γ_c . There is a transition at a Karlovitz number ~ 0.12 above which clear Turing patterns are destroyed.

Sec. 6.3.1. This fraction is plotted in Fig. 6.3 as a function of Ka for different values of the Turing reaction strength γ_c . There is a clear crossover to a region where the concentration difference created by the Turing patterns is less than the cutoff, corresponding to the destruction of the stripes, at a Karlovitz number ~ 0.12 . Although the error bars are large below the transition due to the noisy nature of an active system, there is a reasonable data collapse with no discernable dependence of the position of the transition on γ_c .

These results are for the Schnakenberg model which gives out-of-phase stripes of the A and B concentrations. We have observed that for the Gierer-Meinhardt model, the average amplitude of the pattern collapses when plotted against Karlovitz number for different values of the Turing reaction strength, γ_c (Fig. 6.4), hence the results from the Schnakenberg model do not change significantly. However, we find

that the Gierer-Meinhardt stripes have a much higher concentration difference than the Schnakenberg stripes, and we do not observe the dissolution of stripes in the range of activity parameters that we are able to simulate. The velocity profile is along the edges of the stripes. The stripe concentration profile is irregular even in the absence of activity. There is no regular shear state.

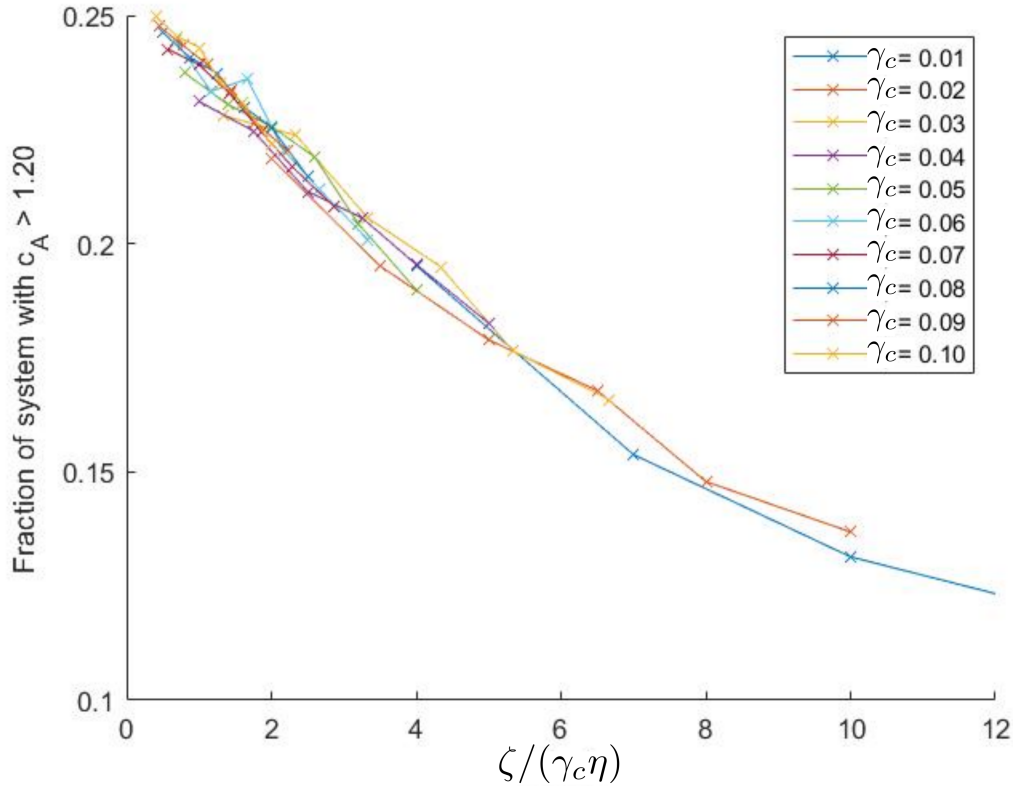


Figure 6.4: Fraction of active Turing system with concentration of species A above a chosen threshold as a function of Karlovitz number for different values of the Turing reaction strength, γ_c - for Gierer-Meinhardt stripes. The data collapses with Karlovitz number, however the stripes do not dissolve in the range of activity parameter that we were able to simulate.

6.3.1 Characterising the dissolution of Turing stripes

In Figs. 6.3, 6.4, we characterized the crossover from Turing stripes to active turbulence by measuring the fraction of the system where the concentration of morphogen A, c_A , is greater than a given cutoff value, finding that the data

collapsed as a function of Karlovitz number. We have checked that the data collapses regardless of the choice of cutoff.

Moreover, we checked that the data collapses for other cutoff-independent parameters - the amplitude of patterns, and the standard deviation of the concentration profile. Fig. 6.5 plots the average amplitude of the Turing patterns for different Karlovitz numbers, once again showing a data collapse. Here, we define the amplitude of a Turing pattern as the difference between the highest concentration and the lowest concentration of c_A in a time snapshot. We average over uncorrelated time snapshots separated 200 lattice Boltzmann timesteps apart to calculate the average amplitude. Small fluctuations in the concentration profile appear as noise in the average amplitude.

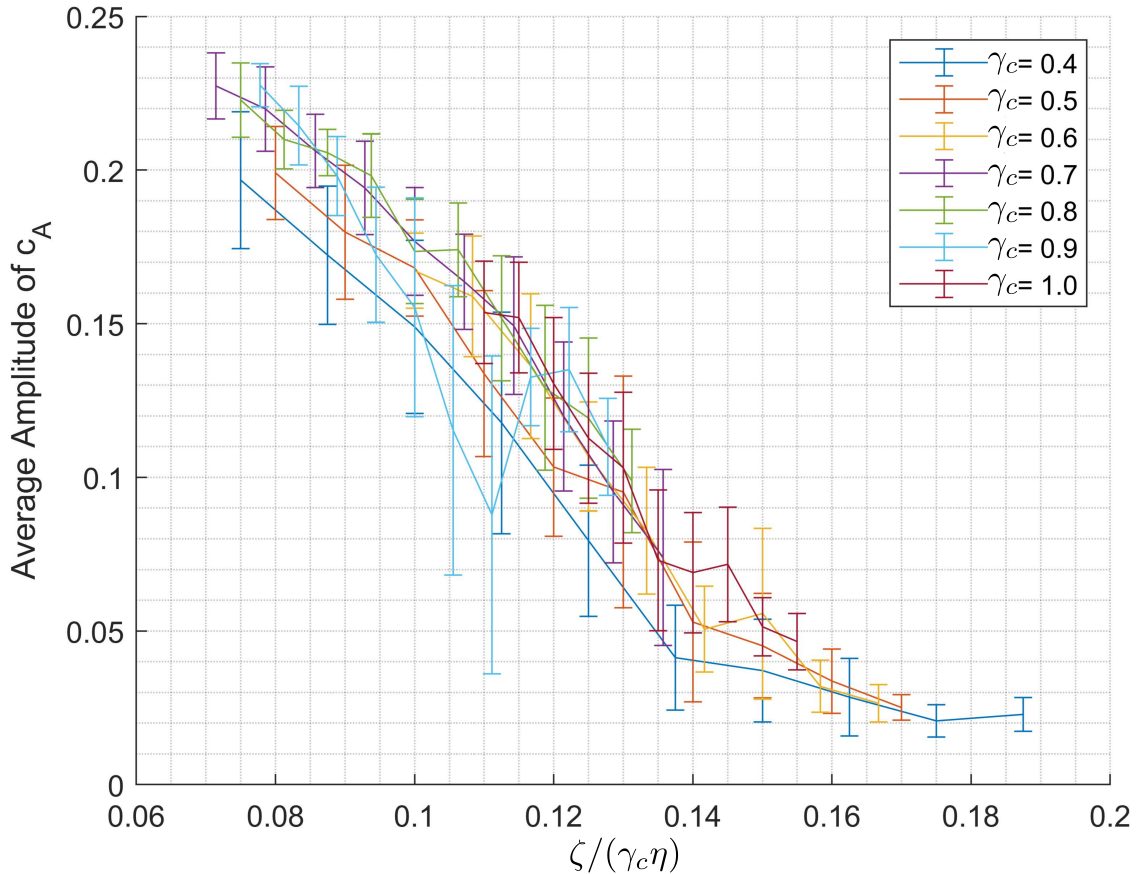


Figure 6.5: Average amplitude of concentration of species A in an active Turing system as a function of Karlovitz number for different values of the Turing reaction strength, γ_c . The amplitude of Turing patterns decreases as a function of Karlovitz number.

6.4 Coupling activity to concentration

In this section, we investigate the effects of allowing the activity to depend on the local value of the morphogen concentrations $\zeta \rightarrow \zeta(c_A, c_B)$. We do this by assigning an activity ζ if $c_A > 1.55$ (equivalently, $c_B < 0.59$) and $\beta\zeta$ if $c_A < 1.55$ (equivalently, $c_B > 0.59$). Here, $c_A = 1.55$ and $c_B = 0.59$ correspond to the mean concentration of the morphogens A and B respectively, which is also the concentration of the morphogen values between the two stripes. Although the reactions themselves are highly non-linear, the emergent concentration fields show a symmetric structure (see Fig. 6.6b). We use an activity parameter $\zeta = 0.07$ for this section.

This assignment of activity is easy to implement numerically for each different region. However, it gives rise to singular force densities at the transition point $c_A = 1.55$, although this is numerically normalized by the finite grid spacing used in our simulations. We also looked at other possible couplings of ζ to c_A , including a linear dependence on c_A , a sinusoidal dependence on c_A , and a case where the discontinuous cutoff is instead smoothed out with a tanh function over 4 lattice sites. In every case, we found similar flow patterns - implying that the results described below are not dependent on the exact functional form of activity, provided that the activity switches from extensile to contractile across stripes.

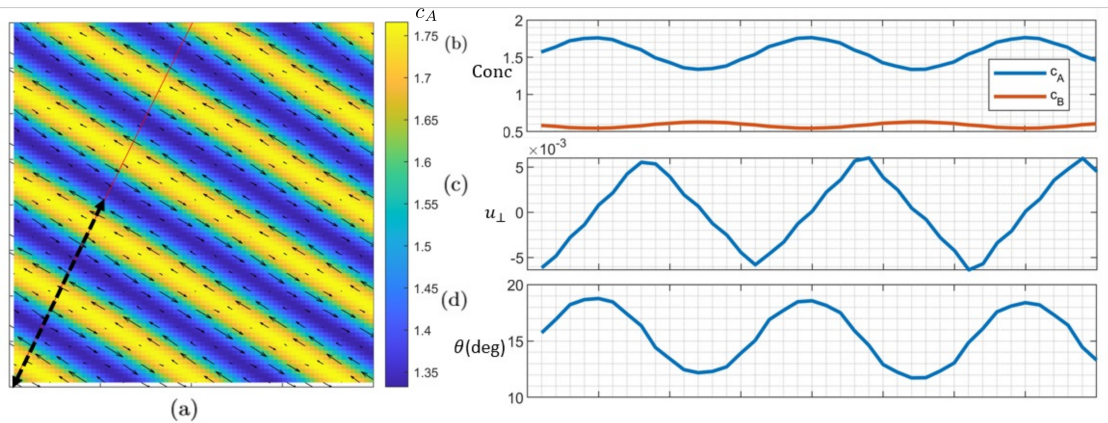


Figure 6.6: Alternating extensile and contractile stripes: (a) Concentration profile of c_A with velocity field marked by arrows. Variation along the black dashed line of (b) concentration fields; (c) velocity perpendicular to the marked line. The shear flow has its highest value at the edge of the stripes; (d) angle of the director with respect to the marked line. The angle is maximum in the extensile region.

First consider the case $\beta = -1$. Regular stripes, with alternating bands of high concentrations of c_A and c_B form at the Turing length scale and span the entire system, as shown in Fig. 6.6a,b. The morphogens respectively generate positive and negative active stresses of equal magnitude. There is Couette flow within each stripe [139], with adjacent stripes having opposite shear (Fig. 6.6a,c).

The angle of the director relative to the normal to the stripe boundaries, θ_0 , is approximately constant, but there is a small superimposed sinusoidal variation with a period equal to the width of two stripes (Fig. 6.6d). The director initially orders at an angle $\theta_0 \sim 45^\circ$ to the normal to the stripes. It then slowly rotates towards $\theta_0 \sim 0^\circ$. When the angle reaches approximately $\theta_0 \sim 10^\circ$ to the stripe, the stripes first bend and then slip along an axis perpendicular to the stripes, at a position determined by the noise. Each stripe breaks at this line and joins with its adjacent neighbour thus increasing the director angle again, and the process repeats (Fig. 6.7).

To understand this behaviour it is helpful to consider first an infinite number of **fixed** stripes of width L with alternating extensile and contractile activities which we take to lie along the y -axis. For a director field at an angle θ_0 to the x -axis the force acting at the boundaries between stripes due to the change in activity is $S_{nem}(\nabla\zeta) \sin(2\theta_0)$ parallel to the stripe edges, and $S_{nem}(\nabla\zeta) \cos(2\theta_0)$ perpendicular to the stripe edges [64]. The forces alternate in sign on successive stripe boundaries and hence lead to alternating shear flows in neighbouring stripes with proportional shear rates $\propto S_{nem}(\nabla\zeta) \sin(2\theta_0)$.

We initiate the simulations with a random noisy director configuration. The director field forms small amplitude sinusoidal oscillations with a wavelength twice the width of a single stripe around a mean angle $\theta_0 \approx 45^\circ$. The extensile stripe has a slightly higher director angle than the contractile stripe.

The dynamics of the director as it responds to the shear field is given by [33, 140, 141]

$$\frac{d\theta}{dt} = \frac{u'}{2}(1 + \lambda \cos 2\theta) \quad (6.8)$$

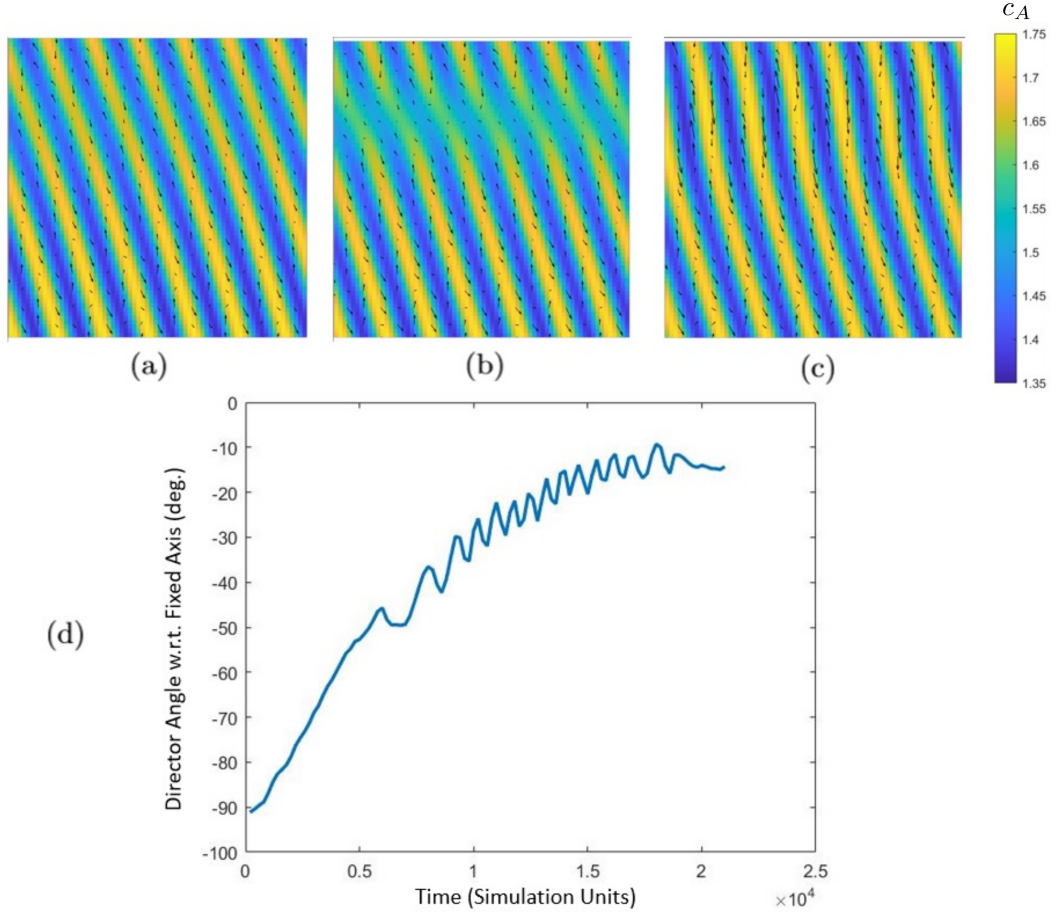


Figure 6.7: Slipping of stripes: (a), (b), (c) sequential time snapshots of a slipping region, taken 200 time steps apart. (d) Variation of the director angle at a point undergoing slipping with time, with respect to a fixed axis. Each slipping event causes the director field to slightly re-set. The superimposed drift at later times occurs because the stripe is slowly rotating.

where u' is the rate of shear across a stripe, and θ is the angle of the director measured from the normal to the stripe at every point in the system. The angle of the director at the stripe edge is given by θ_0 , which sets up the shear flow. Hence, $u' \propto (\nabla\zeta) \sin(2\theta_0)$.

We are considering the flow-tumbling regime, $0 < \lambda < 1$, and hence the first term in the brackets in Eq. (6.8) dominates. Due to the direction of $(\nabla\zeta)$, the shear flow tends to turn the director in contractile stripes towards $\theta = 0^\circ$, whereas rotation towards $\theta = 90^\circ$ is preferred in the extensile stripes. This is indeed the dynamics observed for very small values of the elasticity K . However for larger K this introduces director gradients that are disfavoured by the elastic energy, and

instead the whole system rotates slowly towards $\theta = 0^\circ$.

The reason that rotation towards $\theta = 0^\circ$ is preferred to turning towards $\theta = 90^\circ$ follows from noting that there is a small, approximately sinusoidal, variation of the director field superimposed on its constant background value θ_0 (Fig. 6.6d). This occurs because of the active instability which is then stabilised by the stripe walls [30]. Therefore the director in the contractile stripes takes a slightly lower average value than that in the extensile stripes. Since $\lambda > 0$, it follows from Eq. (6.8) that the rate of rotation is slightly faster in the contractile stripes, and elasticity forces the extensile stripe to follow. As the director rotates, the angle θ_0 becomes lower - this reduces the forces on the boundary ($\sim \sin(2\theta_0)$) and the magnitude of the shear velocity and hence the rotation rate of the director goes down significantly. We note that the angle θ_0 remains fixed if the flow aligning parameter is zero, and the nematic aligns parallel to the stripes if it is negative.

We now relax the pinning of the stripes and return to describe the behaviour of striped Turing patterns with alternating extensile and contractile activities. The reaction-diffusion dynamics promotes each of the Turing stripes to have a fixed width of half the Turing length-scale, given by Eq. (6.5). This width is a function only of the Turing parameters, and any deviation from it induces a strong reactive flux which drives the system back to the Turing length-scale.

Initially, Turing patterns form with the stripe boundaries at an angle of approximately 45° with respect to the director field. At this angle the force at a stripe edge due to the activity gradient is along the stripe edge, and there is no force perpendicular to the edge. This makes it possible for the Turing system to form stripes without an active force breaking them apart. Just as for the fixed boundary stripes the shear flow results in the director angle decreasing towards $\theta = 0^\circ$. This change in director angle creates forces normal to the stripe boundaries. These forces seek to change the width of the Turing patterns, but are balanced by the reaction-diffusion system, which seeks to oppose this change.

As the director angle decreases the normal forces increase and eventually the stripes start to bend at a random position determined by noise. The bending

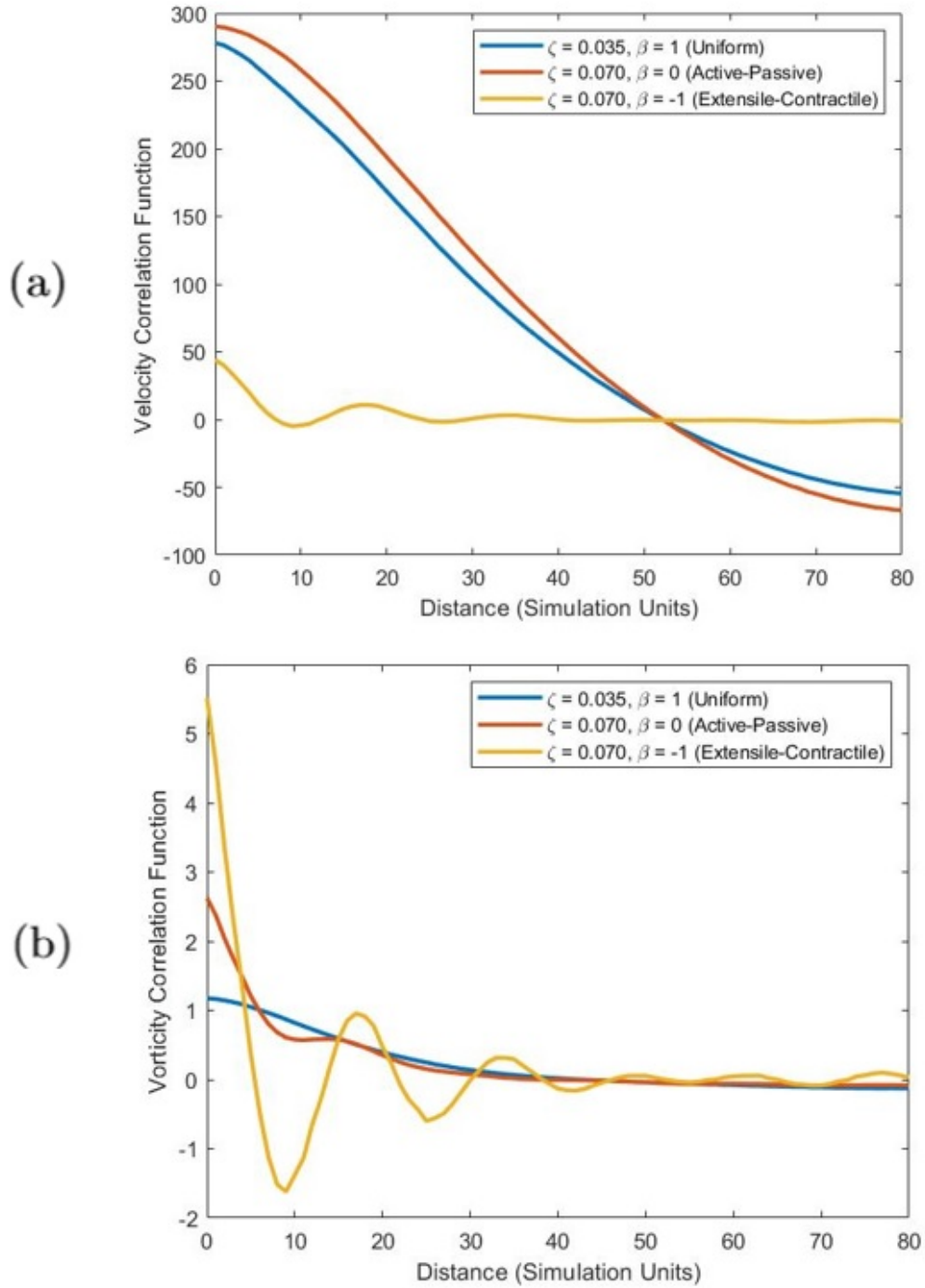


Figure 6.8: Comparison of (a) velocity-velocity and (b) vorticity-vorticity correlation functions for $\beta = 1$, $\beta = 0$, and $\beta = -1$. Non-normalized correlation functions are plotted to contrast the magnitudes of the velocity and vorticity.

increases until the stripes collectively slip by fracturing and connecting to the neighbouring stripe (Fig 6.7a,b,c). The reaction-diffusion dynamics then straightens the stripes. The slipping process increases the director angle with respect to the new stripes. This re-sets the director angle to a higher value and the process repeats (Fig. 6.7d).

For higher activities, the horizontal forces are strong enough to break up the stripes before slipping can re-set the director angle. However, after the stripes break up, the horizontal forces seeking to break stripes apart disappear, and the stripes can re-form in an uncorrelated direction, with the director initially making an angle of 45° with respect to the normal to the new stripes. At still higher activities, we observe patches of morphogens A and B advected by the flow, which are unable to form long stripes.

We have looked at other possible couplings of ζ to c_A , including linear dependence, sinusoidal dependence, and a case where the Heaviside function is instead smoothed out with a tanh function over 4 lattice sites. In every case, we find qualitatively similar flows to the results reported above.

6.5 Active Instability

We recall that the activity is ζ if c_A is greater than a threshold value c_0 and $\beta\zeta$ if c_A is less than c_0 , and hence that the Turing system forms stripes with activities ζ and $\beta\zeta$. In Sec. 3 we considered $\beta = 1$ corresponding to an activity independent of concentration, and in Sec 4 we discussed $\beta = -1$ corresponding to alternating extensile and contractile stripes with activities of equal magnitude. We now consider $\beta = 0$ which leads to alternating active and passive stripes, and then discuss the crossovers as β is varied between these values.

For $\beta = 0$ the behaviour with increasing activity follows the pattern observed for $\beta = 1$, shown in Fig. 6.2. However approximately twice the strength of activity is needed to destroy the stripes for $\beta = 0$ as active energy is only supplied to half the system. To demonstrate this we compare the time-averaged velocity-velocity and vorticity-vorticity correlation functions for $\beta = 1, \zeta = 0.035$, $\beta = 0, \zeta = 0.070$, and

$\beta = -1, \zeta = 0.070$ in Fig. 6.8. The long-range nature of the flow field implies that the velocity correlation function is virtually indifferent to the activity distribution for $\beta = 0, 1$, as shown in Fig. 6.8a. By contrast, the extensile-contractile flow ($\beta = -1$) discussed in Sec. 4 has a different velocity-correlation curve. The magnitude of the velocity is much lower because of the competition between extensile and contractile stresses, and there are oscillations reflecting the underlying stripes. These oscillations appear very clearly in the vorticity-vorticity correlation function (Fig. 5(b)).

We recall that, for $\beta = -1$, the stripes are regular and evolve by slipping along an axis perpendicular to the stripes. Therefore we next allow β to take negative values to understand the crossover from $\beta > 0$ where the existence of the stripes is a competition between the active bend instability destroying the stripes and Turing reaction-diffusion re-forming them.

It is helpful to again first consider an infinite number of **fixed** stripes of width L with alternating extensile and contractile activities which we take to lie along the y -axis. When the extensile activity is significantly stronger than contractile activity, the system forms bend instabilities across the stripes. On the other hand, when contractile activity dominates, the system forms splay instabilities across the stripes. As is well known [62], extensile instabilities are much easier to form than contractile instabilities, and hence an extensile-dominant system forms instabilities faster than a contractile dominated system. We note that when the strength of extensile and contractile activities is approximately equal, the instability formation is very slow - as each type of instability suppresses the formation of the other.

The situation is similar for Turing stripes. However, if extensile and contractile activities are close in magnitude, the angle at the stripe boundary θ_0 can get very low without forming an active instability. If this angle reaches a critical value (approximately 10° with respect to the stripe normal), the stripes start collective slipping behavior, connecting to their next neighbours. This re-sets the director angle to a slightly higher value, and the process keeps repeating (Fig. 6.7d). Therefore a stronger imbalance in the activity is required to form instabilities across Turing stripes than fixed stripes because of the stabilizing slipping behavior.

Once a bend or splay instability has formed across one of the stripes, the instability spreads across the entire system and the shear flow across all the stripes gets replaced by bulk active nematic flow patterns of vortices and active turbulence. This breaks the stripes apart into stripe segments and patches. The director and velocity fields set up in this case are similar to the flow of an active nematic with an effective renormalized activity strength. Given a renormalized effective activity strength all the results from the previous sections carry forward as β increases from -1 once the slipping process (or the balance of activities) fails to overcome the instabilities.

6.6 Conclusions

We have studied the effect of advection by active nematic flows on Turing patterns finding that active flows destabilize Turing stripes. For very low activity, the Turing stripes bend with the flow lines of the active nematic vortices. For intermediate activities, the stripes break up into spots and line segments which are advected by the active nematic flow. For higher activities, the flow destroys the Turing patterns. The point at which this occurs is set by a balance between the active advective flux and the Turing flux.

We then introduced a coupling between the activity of the nematic and the local concentration of the morphogens. In the case where alternating stripes of morphogens have activity of opposite sign (extensile and contractile) but equal strength, the stripes form a series of alternating shear flows across the system, with the highest flow achieved at the boundary between two stripes. We observe numerically that the stripes collectively fracture and slip sideways to join their neighbours on long timescales, which stabilizes the stripe system against bend or splay instabilities.

Phase-separated stripes can also be obtained using mechanisms other than Turing patterns - for instance, similar stripes have been studied in two-dimensional mixtures of an active polar gel and passive isotropic fluid, with an emulsifying surfactant [142]. The flow fields we observe when the activity depends on concentration

are reminiscent of the flows observed in polar lamellar systems with contractile activity [142] or external forcing [143].

If the activity in all stripes is of the same sign, or if one of the alternating stripes has significantly higher activity than the other, instabilities form across the stripes. These are bend instabilities if extensile activity dominates, and splay instabilities in a predominantly contractile system. These instabilities destroy the striped Turing pattern in favour of spots and line segments. The resulting patches of activity set up flows analogous to a bulk active nematic, but with a reduced activity.

In this chapter, we have considered only the case where the lengthscale of patterning is comparable to the active lengthscale. It would be interesting to study the case of Turing spots, when the lengthscale of patterning is much larger than the active lengthscale. In this case, we may expect to see the effects of anchoring at the spot boundaries, and also see different active flows in different regions. In addition, studying this system in a flow-aligning regime might lead to different flow states.

While we did not model any particular biological system, our results shed light on the interplay between mechanical active stresses and chemical patterning, and highlight the possibilities of new dynamical states arising from mechano-chemical feedback loops. Understanding this interplay between mechanical stresses and chemical patterning is important to studying development and morphogenesis [144–146].

Take-home message:

The balance between chemical patterning and active flows can be quantified by a Karlovitz number. Coupling chemical patterns and active matter can lead to new flow states with mechano-chemical feedback.

“Coming back to where you started is not the same as never leaving.”

—Terry Pratchett, *A Hat Full of Sky*

7

Active Matter under Viscoelastic Confinement

In this chapter, we look at mechanical feedback between active nematics and their confinement by numerically studying an active nematic channel confined by viscoelastic walls. We find that this system exhibits periodic flow reversals over a range of activity and elasticity, and explain the mechanism underlying these oscillations. This is an example of how mechanical feedback with external confinement can alter the behaviour of active matter.

7.1 Introduction

In addition to biochemical cues, mechanical interaction with the environment plays an important role in dictating the collective motion of living systems. For instance, cells migrate towards stiffness gradients in the substrate, in a phenomenon known as durotaxis [147]. Interaction with the surroundings plays an important role for bacterial colonies in a polymeric matrix [148–150], or the migration of cells through an extracellular matrix [15, 151–153]. In a more synthetic setting, traction force microscopy is used to measure cellular force fields using an elastic substrate [154, 155], however, this technique does not account for how cellular behaviour might change due to forces from the substrate itself. Therefore, understanding

the interplay between the elasticity of the confining medium and active cell layers is crucial to studying mechanobiology.

It is well known that active nematics confined inside a rigid channel display a spontaneous flow transition —transitioning from quiescence to laminar flow — when the activity is increased beyond a threshold value [30, 156–159]. Increasing the activity further induces a cascade of dynamical transitions resulting in oscillatory flows [160, 161], dancing topological defects [106, 161], and active turbulence [112, 162–165]. Active nematics in channels are a paradigmatic system to study confined active matter [166]. Therefore, it is timely to investigate the interaction between activity and viscoelasticity, by analyzing an active nematic confined by a soft viscoelastic channel.

Previous research that addresses the role of viscoelasticity in living systems has looked at active particles within a viscoelastic fluid [167–172] or active matter in contact with a viscoelastic environment [173–175]. In the latter, less studied, case numerical simulations have demonstrated that temporal pulses in activity drive reversal of spontaneous flows [174]. Moreover, spontaneous flow reversals have been experimentally observed in swimming bacteria confined in a disk [176]. In this chapter, we demonstrate numerically that, above a critical activity, soft viscoelastic confinement produces spontaneous flow states in an active nematic that switch flow directions periodically. The direction-reversing oscillatory flows disappear when the elastic modulus of the confinement increases above a critical value. Building on our findings, we explain the origin of oscillations as the interplay of activity and viscoelasticity, demonstrate the generality of the phenomenon, and discuss the consequences.

The remainder of the chapter is organized as follows. In Sec. 7.2, we describe the equations describing an active nematic confined by soft viscoelastic walls. We then introduce two possible constitutive relations for the viscoelastic material and discuss how to simulate this system numerically. In Sec. 7.3, we use this model to investigate spontaneous flow reversals in viscoelastic channels. We discuss the mechanism underlying these and show that the spontaneous flow reversal state

is the first unstable mode for a large range of activity and elasticity. Finally, in Sec. 7.4, we conclude with a summary of our results.

7.2 Setup

We consider a two-dimensional channel of width $2L$ along the y -axis containing an active nematic fluid. The channel is bordered by viscoelastic slabs on both sides, each spanning a width of $(\beta - 1)L$. The other end of the viscoelastic material is attached to a rigid wall, with no-slip boundary conditions. This setup is shown in Fig. 7.1, with the blue, yellow and black regions showing the active nematic channel, viscoelastic confinement, and rigid wall respectively. We choose the origin $y = 0$ in the centre of the channel.

The active nematic is modelled by an orientation field $\mathbf{Q}$ and velocity field $\mathbf{u}$, described by the equations of nematic hydrodynamics (Eqs.(2.12), (2.2)). For simplicity, we consider strong planar anchoring of the director field at the interface, i.e., $\theta(\pm L) = 0$.

The dynamics of the viscoelastic slabs are described using a displacement field $\mathbf{w}$, describing the displacement of material from its equilibrium position. This displacement field obeys the equations of motion [177, 178]

$$\nabla \cdot \mathbf{w} = 0, \quad (7.1)$$

$$\rho_2 \frac{\partial^2 \mathbf{w}}{\partial t^2} = -\nabla P_2 + \nabla \cdot \boldsymbol{\tau}, \quad (7.2)$$

where ρ_2 is the gel density, P_2 is the bulk pressure in the viscoelastic layer, and $\boldsymbol{\tau}$ is the stress tensor inside the viscoelastic slabs. The viscoelastic material is in contact with a no-slip wall at $y = \pm\beta L$, thus

$$\mathbf{w}(\pm\beta L) = 0. \quad (7.3)$$

The stress tensor $\boldsymbol{\tau}$ is model-dependent and encodes the constitutive relations of the viscoelastic material. In this chapter, we study the constitutive relations for (i) elastic confinement and (ii) viscoelastic confinement using the Maxwell model, the dynamics of which are discussed below.

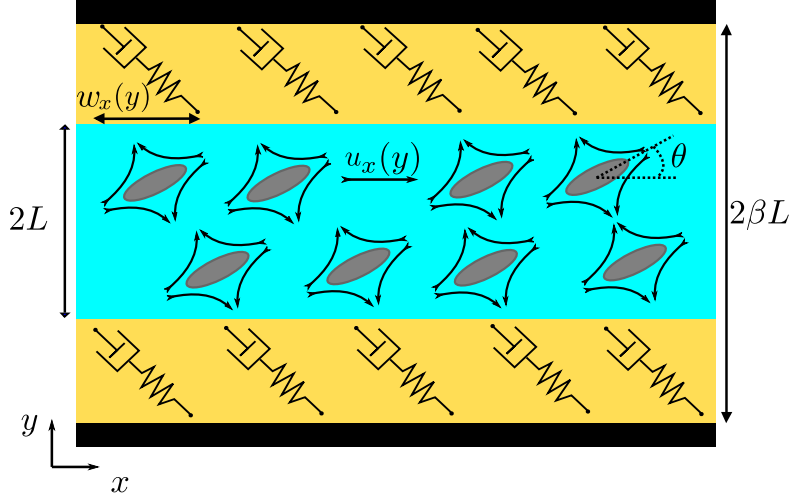


Figure 7.1: Schematic representation of the system: an active nematic layer of width $2L$ (shown in blue) is confined between two viscoelastic layers, each of width $(\beta - 1)L$ (shown in yellow). The width of the entire setup is $2\beta L$. The active nematic has a velocity field $\mathbf{v}$ and orientation field θ . The viscoelastic layers have a displacement field $\mathbf{u}$.

7.2.1 Modelling the Confining Material

In this section, we discuss the constitutive equations for a solid with linear elasticity and a viscoelastic fluid which is solid-like at short timescales, and liquid-like at long timescales.

Elastic Confinement

In purely elastic confinement, we have the constitutive relation [179]

$$\boldsymbol{\tau} = E \left(\nabla \mathbf{w} + (\nabla \mathbf{w})^\top \right), \quad (7.4)$$

where E is the shear elastic modulus of the confinement. This is the familiar form of linear elasticity, where the stress is proportional to the strain through an elastic coefficient.

Furthermore, at the interface between the active nematics and the viscoelastic layer, we impose no-slip conditions,

$$\mathbf{u}(\pm L) = \partial_t \mathbf{w}(\pm L), \quad (7.5)$$

and continuity of the stress tensor,

$$\boldsymbol{\Pi}(\pm L) = \boldsymbol{\tau}(\pm L). \quad (7.6)$$

where $\mathbf{\Pi}$ is the usual stress tensor inside the nematic channel (Eq.(2.20)). We also assume instantaneous relaxation inside the viscoelastic i.e. $\rho_2 = 0$ in Eq.(7.2).

We numerically solve the active nematic equations using a hybrid Lattice-Boltzmann method, for a channel with free-slip and no-penetration boundary conditions. The boundary layer of the nematic fluid drags the boundary of the elastic material, and the elastic material exerts an elastic body force on the boundary layer of the nematic. Inside the elastic material, the fields are solved on a rectangular lattice using the Euler method, approximating derivatives using a finite difference scheme. This involves iteratively solving Eqs.(7.1)-(7.2) on a fixed grid with the boundary conditions (Eqs.(7.3), (7.5),(7.6)) imposed at every LB time step.

Viscoelastic Confinement

We model viscoelastic confinement by using the constitutive relations for the Maxwell model [180]

$$\frac{1}{E} \overset{\nabla}{\boldsymbol{\tau}} + \frac{1}{\eta} \boldsymbol{\tau} = \nabla \partial_t \mathbf{w} + (\nabla \partial_t \mathbf{w})^\top. \quad (7.7)$$

In this model, $\overset{\nabla}{\boldsymbol{\tau}}$ is the upper convected derivative [178] required to preserve Galilean invariance, and E and η are the shear elastic modulus and viscosity respectively. A Maxwell material can be thought of as a spring and a dashpot connected in series. It behaves as an elastic solid at short times and as a viscous liquid at long times, with the crossover timescale η/E . The viscoelastic material again satisfies no-slip conditions at the rigid wall (Eq.(7.3)), and continuity of stress at the interface (Eq.(7.6)).

There are many other choices of viscoelastic models- notable ones include the Kelvin-Voigt model [181, 182], the Olroyd-B and Olroyd-A models [183], and viscoelasticity models involving fractional derivatives [184]. These models all capture a wide range of different viscoelastic flow and creep behaviours, showing different short term and long term behaviours. In this chapter, we only consider the upper-convected Maxwell model as one of the simplest models of viscoelastic fluids.

We simulate this model using a phase-field φ , which is described in Sec. 2.6. The boundary between the phases was allowed to relax to an free-energy minimum (Eq.(2.22)) to create a smooth interface, and then the field φ was kept fixed for the simulation. Since we used a smooth boundary between the two phases, we defined the stress tensor in the transition region $0 < \varphi < 1$ as the weighted average of the stress tensors in each region

$$\mathbf{\Pi}^{net} = \varphi \mathbf{\Pi} + (1 - \varphi) \mathbf{\tau} \quad (7.8)$$

where $\mathbf{\Pi}^{net}$ is the stress tensor that we use in Eq.(2.2).

We numerically solve the active nematic and Maxwell model equations using a hybrid Lattice-Boltzmann method, with the dynamics of the stress tensor $\mathbf{\tau}$ (Eq.(7.7)) solved using a finite-difference method.

For our simulations, we use the parameter values $\eta = 10/3$, $\gamma = 10$, $\rho_1 = 20$, $\rho_2 = 0$, $K = 0.1$, $S_{nem} = 0.25$, $L = 10$, $\lambda = 0$, and $\beta = 2$, run over 200,000 LB timesteps. The simulations are initialized with a tiny perturbation ($\sim 10^{-3}$ rad) in the lowest excited state of the orientation field.

7.3 Spontaneous Flow Reversals in Elastic and Viscoelastic Channels

We first consider a purely elastic material bounding the nematic fluid. In the limit of large elastic modulus $E \rightarrow \infty$, the boundaries at $y = \pm L$ are rigid and we recover the classical result of Voituriez et al [30]: increasing the activity beyond a critical value $\zeta_{wall}^c = -\frac{\pi^2 K [\eta/\gamma + 2S_{nem}^2(1-\lambda)^2]}{S_{nem}(1-\lambda)L^2}$, we find spontaneous bidirectional flows inside the channel driven by the distortions in the director field. In the opposite limit $E = 0$, corresponding to a film of an active nematic in viscous confinement, an analogous transition to a steady flow is observed at activity $\zeta_{free}^c = \zeta_{wall}^c/4$, also first reported by Voituriez et al. [30].

The phase diagram of the system for different values of activity and elasticity is shown in Fig. 7.2. The different flow states obtained from numerical simulations are shown as symbols. The critical activity of the system at intermediate values of

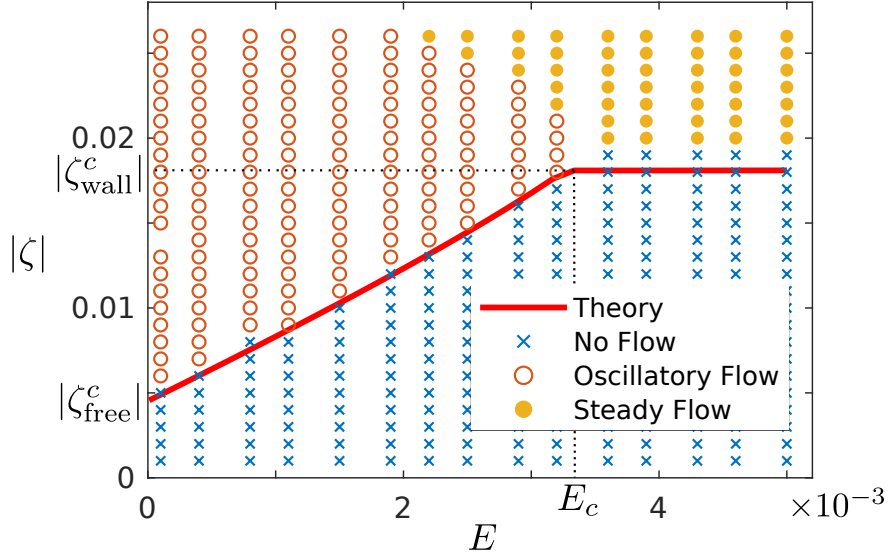


Figure 7.2: Phase diagram in the (ζ, E) plane, illustrating the different states of an active nematic when confined in a channel with elastic walls. The continuous red line is the critical activity ζ^c , obtained from linear stability analysis [3], at which the nematic state becomes unstable driving flows. The symbols show the different states obtained from numerical simulations after 200,000 timesteps, and represent no flow, periodic oscillations and steady flow states. Above a critical elasticity E_c , the confinement starts to behave like a rigid wall and we recover the steady flow state.

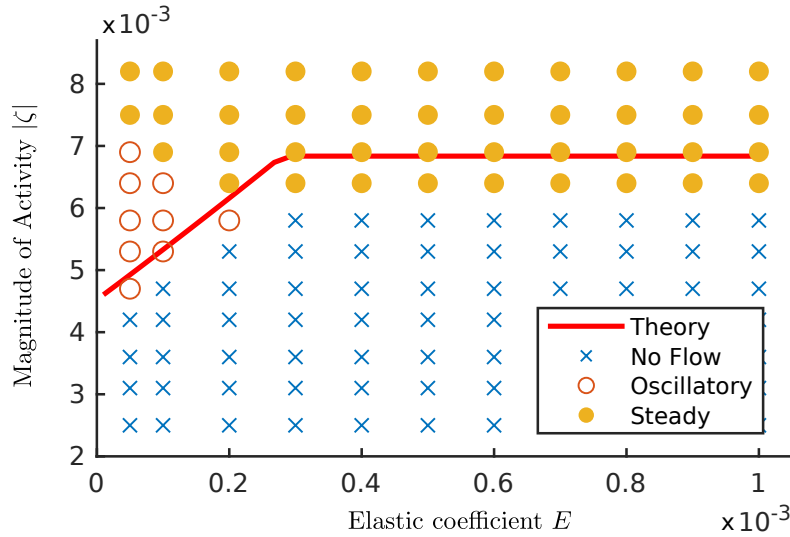


Figure 7.3: Phase diagram in the (ζ, E) plane, illustrating the different states of an active nematic when confined by viscoelastic walls described by a Maxwell model. The continuous red line is the critical activity ζ^c , obtained from linear stability analysis [3], at which the nematic state becomes unstable driving flows. The symbols show the different states obtained from numerical simulations after 200,000 timesteps, and show no flow, periodic oscillations and steady flow states. Above a critical elasticity E_c , the confinement starts to behave like a rigid wall and we recover the steady flow state. The region of parameter space showing the flow reversal state is much smaller for the viscoelastic case.

E , as derived from linear stability analysis, is shown by the red line. The critical activity $\zeta^c = \zeta_{\text{free}}^c$ at $E = 0$, and increases with an increase in the elastic modulus E , until a threshold elastic modulus $E = E_c$. Beyond E_c the critical activity “freezes” to $\zeta^c = \zeta_{\text{wall}}^c$, that corresponds to a rigid wall.

The first stable flowing state is different for $E < E_c$ and $E > E_c$. For $E < E_c$ the active nematic transitions from a quiescent state to a bi-directional flow state with the flow direction reversing periodically. On the other hand, for $E > E_c$, the bi-directional flow state is stable and no oscillations are observed. The oscillatory state persists for higher activities above the critical flow transition.

We find qualitatively similar oscillations when the confinement is described by the Maxwell model. However, the range of activity and elasticity where this periodic flow state is observed is smaller, as seen in Fig. 7.3. Unless the oscillations are very fast, the viscous flows inside the Maxwell material dissipate the elastic energy stored inside the confinement, and oscillations die out to yield the steady flowing state.

In Figs. 7.2 and 7.3, we find a small discrepancy between the numerical and the analytical solution. We attribute this to two possible sources of error.

First, we expect discretization errors from a first order finite difference scheme, and spurious velocities due to Lattice-Boltzmann [52, 185, 186]. This is especially true at the boundary, where there is positive feedback between the velocity and the displacement field. This error can be controlled by changing the timestep of the simulation, rescaling the simulation parameters to use a smaller lattice spacing, or using higher order finite difference schemes.

Second, we expect discretization errors due to boundary localization. In the case with elastic boundaries, the LB channel boundary conditions are not implemented exactly on the boundary nodes, but rather between the boundary node and the wall node. On the other hand, the displacement field and restoring force are coupled to the boundary node. This error can be controlled by rescaling the simulation parameters to use a smaller lattice spacing, since it depends on the discretization of space Δx .

7.3.1 Mechanism of Flow Reversals

The oscillatory state can be understood by following the temporal evolution of a system in the periodic flow reversal regime. This can be described in four phases:

I. At the start of the cycle (see Fig. 7.4a), the elastic layer is not deformed ($u_x = 0$), and the stress at the active nematic-elastic interface ($y = \pm L$) vanishes. This condition corresponds to a free-standing active nematic channel, with no resistance from the elastic layer, which has spontaneous flows for activities above a critical activity $|\zeta| > |\zeta_{\text{free}}^c|$.

II. The flows in turn deform the elastic confinement through no-slip boundary conditions at the edge. The confinement produces an elastic response to the deformation, which results in a retarding force acting on the edges of the channel through the continuity-of-stress boundary condition (see Fig. 7.4b).

III. Eventually, the elastic response of the channel wall slows down the flow sufficiently, and the flow velocity at the active-elastic interface vanishes (see Fig. 7.4c). At this point, the elastic material has only been strained in one direction over the course of this cycle and has stored elastic potential energy in the displacement field. At this point, the fluid layer at the boundary (which is at rest) experiences internal active forces and an opposing elastic force from the confinement.

IV. If the elastic force overcomes the active force, the resultant force pushes the active nematic fluid in the opposite direction, leading to a flow reversal (Fig. 7.4d). The displacement field of the elastic material continues relaxing to its rest state, imparting energy to the flow in the channel. When the elastic field is relaxed, there is a flow in the channel in the opposite direction and the whole cycle repeats.

Whether the system undergoes spontaneous flow reversals or steady flow depends on the balance of active and elastic forces at configuration III, which has been worked out theoretically in Ref. [3]. The flow in the channel is steady for high elasticity (consistent with the $E \rightarrow \infty$ limit corresponding to a rigid wall), and the flow reversals are observed as the steady state for walls with a lower elastic coefficient. The period T of the oscillations is set by the elasticity of the confinement, viscosity of the channel, and the relaxation timescale of the director field $L/(\gamma^{-1}K)$ [3].

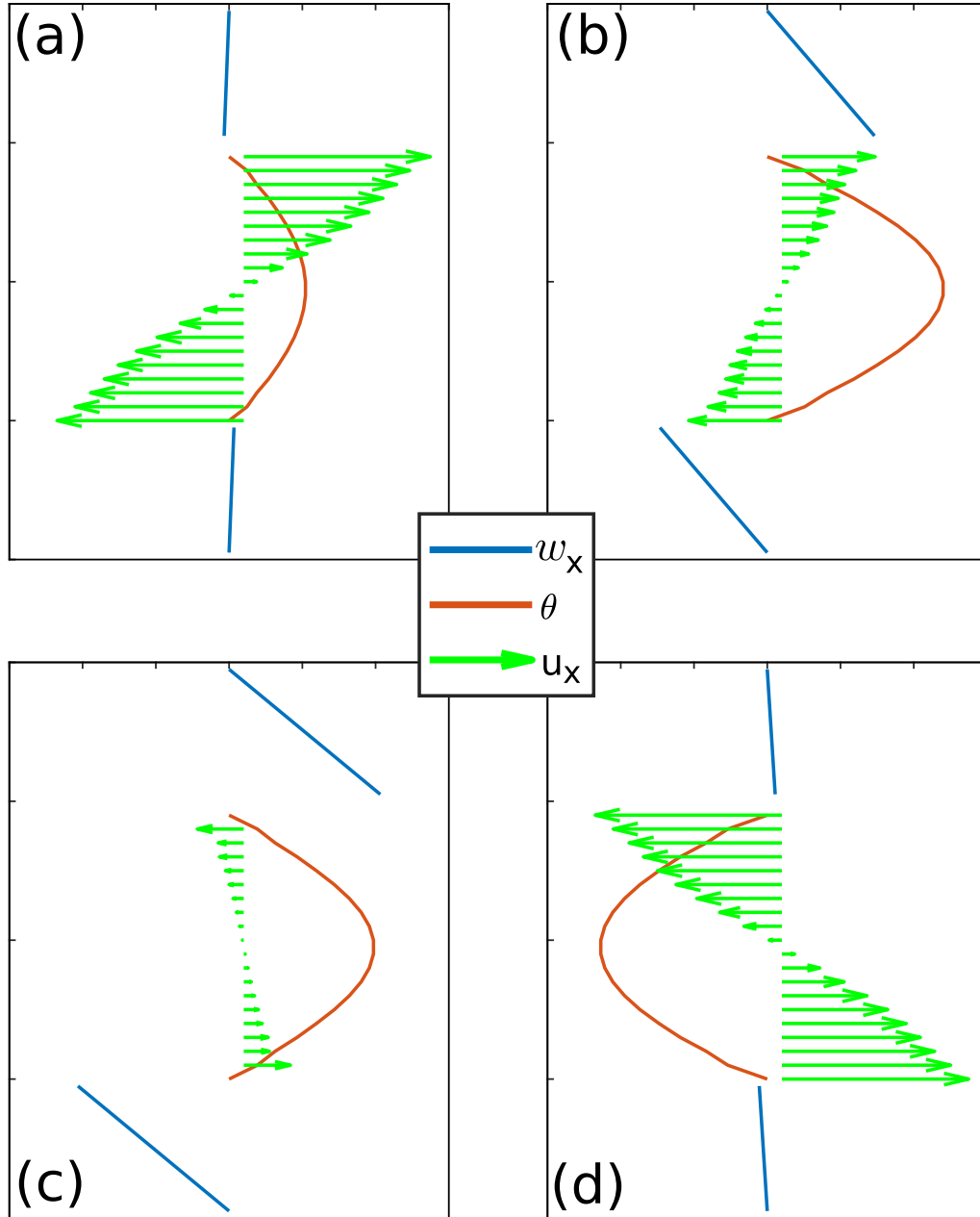


Figure 7.4: Numerical data showing the mechanism of elastic oscillations. Blue, green and red curves show the elastic deformation, channel flow velocity, and nematic orientation respectively. (a) The elastic layer is undeformed, and the channel has spontaneous bidirectional flows. (b) The elastic layer is dragged with the flow field, and exerts restoring forces which retard the velocity field. (c) The flow in the channel flips direction due to the elastic forcing. Now, the elastic field starts to relax back to equilibrium. (d) The elastic layer has returned to the undeformed state, but the flow in the channel is in the opposite direction. The system goes through the same process again.

7.4 Discussion

In this chapter, we studied the mechanical feedback between active nematics and their confinement. We first introduced two different constitutive relations for the material of the confining walls - first the model of linear elasticity, and then the viscoelastic Maxwell model. We also introduced the phase-field equations required to model this system numerically. Using simulations, we showed that active nematics in elastic or viscoelastic confinement show spontaneous flow reversals, and investigated the mechanism of these flow reversals. We found that a spontaneous flow state with periodic reversals is the stable configuration for a large range of activity and elasticity.

Our results highlight confinement as a pathway to spatiotemporal pattern formation in active matter. Remarkably, time-periodic oscillatory flows arise at constant activity. Our predictions can be tested experimentally on confining cell layers or microtubule-motor protein in channels with soft walls. Moreover, traction force microscopy may serve as a potential platform to study the role of mechanical interactions between active cell layers and an elastic substrate.

Take-home message:

Active materials in viscoelastic confinement can lead to spontaneous oscillations.

“Everything should be made as simple as possible, but not simpler.”

— Albert Einstein

8

Modelling Cancer Tumours as Confined Active Droplets

We model malignant breast cancer tumours inside a viscoelastic extracellular matrix (ECM) as a confined active nematic in this chapter. We find that active droplets continually split and merge, giving rise to a dynamical steady state which accurately reproduces the sizes and shapes of cancer tumours obtained from patient data. We then discuss the effects of active flows on the confining ECM - in particular, looking at defect densities and the orientation of ECM fibres at the edge of cancer clusters, and correlate this information with patient data. We conclude this chapter by showing that motility, instead of growth, dominates the dynamics of these tumours. Our results highlight the potential of applying active matter physics to understand the physics of cancer migration and metastasis.

8.1 Introduction

Understanding how cancer cells migrate collectively remains a significant hurdle in mechanobiology. Malignant cancer cells can travel together, maintaining cell-to-cell connections with their neighbours. The details of this coordinated movement are very complex and depend on both the cell type and also the local microenvironment [15]. A better understanding of the mechanisms underlying metastasis could lead to

more accurate ways to predict cancer relapse, and lead to the development of new techniques to control cancer progression. In this chapter, we show that malignant breast cancer tumours inside an extracellular matrix (ECM) can be modelled as an active nematic droplet under viscoelastic confinement.

The remainder of the chapter is structured as follows. In Sec. 8.2, we describe the system of study, writing down the constitutive equations of the cancer tumour and the ECM, and describing the method of simulating this system using a phase-field approach. We choose simulation parameters by inferring them from experimental data and making order of magnitude estimates. In Sec. 8.3, we test the predictions of this model with the sizes and shapes of cancer tumours obtained from clinical samples. We find very good agreement between clinical data and simulations over a range of cluster sizes and discuss some of the assumptions and approximations necessary to arrive at this result. We investigate the fingerprints of activity in the ECM in Sec. 8.4, focusing in particular on topological defects and the anchoring of the ECM at the boundary of the cancer cluster. These are important markers that can be measured in clinical images, which can then be used to infer properties of the dynamical behaviours of the system. In Sec. 8.5, we argue using order-of-magnitude estimates that motility, not growth, is the dominant activity in the system. We conclude this chapter by summarizing our results in Sec. 8.6.

8.2 Breast cancer tumours inside the ECM

Breast cancer happens when cells in the breast tissue mutate and begin to proliferate uncontrollably [187, 188]. The most common type of breast cancer is ductal carcinoma, where epithelial cells in the milk ducts divide and form a tumour mass. Tumours can be benign, and remain constrained by a basement membrane embedded in the ECM. These are called Ductal Carcinoma in situ (DCIS). However, some cancer tumours break free of the basement membrane and spill over into the ECM, forming many malignant cancerous tumours [188, 189]. A clinical sample showing malignant ductal carcinoma is shown in Fig. 8.1a (adapted from Ref. [190] due to privacy reasons).

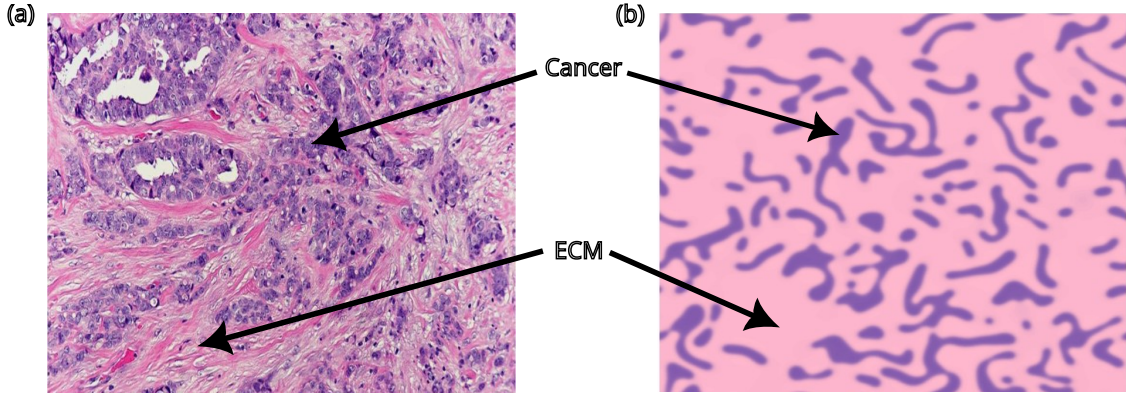


Figure 8.1: Malignant cancerous tumours in ECM: (a) Clinical sample, adapted from Ref. [190], showing malignant ductal carcinoma embedded inside ECM. (b) Snapshots showing many small simulated active cancer clusters in a viscoelastic ECM. In both panels, cancer cells are shown in magenta and the ECM is shown in pink.

Our results are based on tissue image data of these malignant tumours taken from $N = 2012$ invasive breast cancer patients. Each patient was represented by a tissue core of diameter $\approx 1\text{mm}$ sampled from within the malignant tissue, which was then stained and digitized. The digital images were subsequently processed to obtain the sizes and shapes of cancer cluster cross-sections, and the orientation of fibres inside the ECM. This clinical data was obtained in collaboration with Prof. Axel Niendorf at Pathology Hamburg-West, and analyzed by P. Gottheil and Prof. J.A. Käs at Leipzig University.

8.2.1 Modelling

Cancerous cells in malignant breast tumours are elongated in shape, and the cells exhibit nematic ordering [103] inside the clusters. Moreover, cancer cells exert forces on their surroundings, both due to cell growth and motility. We model cancerous tumours as active nematics, with an orientation field $\mathbf{Q}^c$ and velocity field $\mathbf{u}$ coupled to each other by the equations of nematic hydrodynamics (Eqs.(2.12), (2.2)). The total stress in this region is given by

$$\mathbf{\Pi}^c = -p\mathbf{1} + \mathbf{\Pi}^{visc} + \mathbf{\Pi}^{el} + \mathbf{\Pi}^{act}, \quad (8.1)$$

which is the familiar form for an active nematic fluid (Eq.(2.20)) with viscous terms, nematic backflow terms, and active terms respectively.

The ECM is a soft solid with complex rheological properties [191]. While many different viscoelastic constitutive relations have been proposed in the literature to model the ECM, in this chapter we use the Kelvin-Voigt model as the simplest model for a soft solid.

A Kelvin-Voigt material [181, 182, 192] satisfies a constitutive relation given by

$$\mathbf{\Pi}^{visc-el} = -p\mathbf{1} + 2\rho\nu \mathbf{E} + E \mathbf{T}, \quad (8.2)$$

where p is an isotropic pressure term chosen to ensure incompressibility, ν is the kinematic viscosity, E is the elastic modulus, $\mathbf{E} = [(\nabla \mathbf{u}) + (\nabla \mathbf{u})^T]/2$ is the strain rate tensor and $\mathbf{T} = \int \mathbf{E}(t) dt$ is the strain tensor. This model is the simple Kelvin-Voigt model. In general, one can use a generalized Kelvin-Voigt model to keep track of convected and co-rotated terms [55, 192], and also to model more complex materials [192].

In our simulations, we have checked post-facto that the timescale of the elastic restoring term $\tau_{el} \approx 700$ LB (obtained from calculating the time-correlation of T_{yx}) is faster than the timescale of the advective term $1/\left|\frac{u_k \partial_k T_{yx}}{T_{yx}}\right| \approx 1850$ LB, even for intermediate and higher activities. While the elastic restoring term is dominant, the magnitude of the advective term is large enough to affect the details of individual simulations. Despite this, we will find that our simulation results agree well with clinical data, possibly because of the robustness of our results to a change of parameters after averaging over a range of activities (Sec. 8.3.3).

The ECM is comprised of collagen fibres at high densities which form a nematic phase. We model the ECM orientation by a nematic tensor $\mathbf{Q}^e$, which is coupled to $\mathbf{v}$ by the equations of nemato-hydrodynamics (Eqs.(2.12), (2.2)). The total stress in the ECM is therefore given by

$$\mathbf{\Pi}^e = \mathbf{\Pi}^{visc-el} + \mathbf{\Pi}^{el}, \quad (8.3)$$

where we have added the usual term $\mathbf{\Pi}^{el}$ to account for the nematic backflow term in the ECM.

To solve this model numerically, we use a phase field φ (Sec. 2.6) to describe the different regions of the simulation domain, where $\varphi = 1$ represents cancer clusters, and $\varphi = 0$ represents the viscoelastic ECM. This phase field has an associated free energy given by Eq.(2.22), and its dynamics are governed by a Beris-Edwards equation given by Eq.(2.23). The free energy term in φ also generates capillary stress in the underlying fluid [65]

$$\mathbf{\Pi}^{cap} = \left(f_\varphi - \frac{\partial f_\varphi}{\partial \varphi} \varphi \right) - \nabla \varphi \left(\frac{\partial f_\varphi}{\partial (\nabla \varphi)} \right). \quad (8.4)$$

The effect of the capillary stress is to model the stresses generated in the fluid due to surface tension, which accounts for the capillary pressure arising at an interface between different phases [65, 67]. We have ignored this capillary stress term in previous chapters, because the phase field φ was fixed to very regular shapes. The capillary pressure arising from surface tension would simply offset the magnitude of isotropic pressure for circular droplets (Ch. 5), and have no effect on a rectangular channel with no curved boundaries (Ch. 7).

Finally, to account for the soft boundary in the simulations, we interpolate the stress tensor over the transition region $0 < \varphi < 1$ by taking the weighted average of the stress tensors in the intermediate region. The total stress tensor acting on the fluid is thus

$$\mathbf{\Pi} = \varphi \mathbf{\Pi}^c + (1 - \varphi) \mathbf{\Pi}^e + \mathbf{\Pi}^{cap}, \quad (8.5)$$

which drives flows in the fluid according to Eq. 2.2. This formulation, with a smooth interface, also ensures continuity of stress across the interface. We verify using simulations that our interface is stable with our chosen parameters in the absence of activity.

We solve the system of equations using a hybrid Lattice-Boltzmann method on a periodic grid. This involves solving the flow field (Eqs.(2.1) and (2.2)) using a Lattice-Boltzmann method, and the remaining equations (Eqs.(2.12) and (2.23)) using a finite-difference algorithm.

8.2.2 Mapping of parameters

In this sub-section, we discuss how to estimate parameters from the clinical samples to use in our model of cancer clusters.

Elastic and viscous moduli:

We estimate the order of magnitude of the viscous G'' [Pa] moduli within cancer clusters and ECM via frequency-dependent AFM measurements on a primary human breast cancer sample from the University Hospital in Leipzig. At low frequencies ($f = 0.01\text{Hz}$), the viscous moduli for both cancer tumours and ECM is around $G'' \sim 10 \text{ Pa}$, resulting in an effective viscosity of $\eta_1 = \eta_2 = 160 \text{ Pa} \cdot \text{s}$ for the entire system. Cancer clusters fluidize the ECM in their vicinity [193, 194], making it hard to accurately measure the elastic shear modulus in the system. Typical scales of the ECM shear elastic modulus can be estimated between 100 Pa - 1 kPa [195]. For a breast cancer sample measured with AFM, we estimate a shear modulus of the ECM $\sim 400 \text{ Pa}$.

Lengths and velocities: Typical cell motility velocities are measured to be roughly $v_0 \approx 3\mu\text{m}/h$ in vital primary human breast cancer [196]. Cancer cell tumours range over a wide variety of lengthscales, but typical cluster radii are on the scale of $R \sim 10 - 100\mu\text{m}$.

Surface tension: We estimate this roughly from cultured breast cancer cell spheroid fusion experiments reported in Ref. [197].

Active forces: Activity models the typical forces exerted by the cancer cells on their environment [103], which can include traction forces due to motility, and forces due to proliferation. Recent work has shown that these traction forces play an important role in cancer metastasis [198]. The typical magnitude of these forces is around $\zeta \sim 400 \text{ Pa}$ [199], which is comparable to the shear elastic modulus of the ECM.

With these scales, we can set normalized simulation units, hereafter referred to as ‘LB units’. We choose the typical length, time and stress units to be $\Delta x = 5\mu\text{m}$,

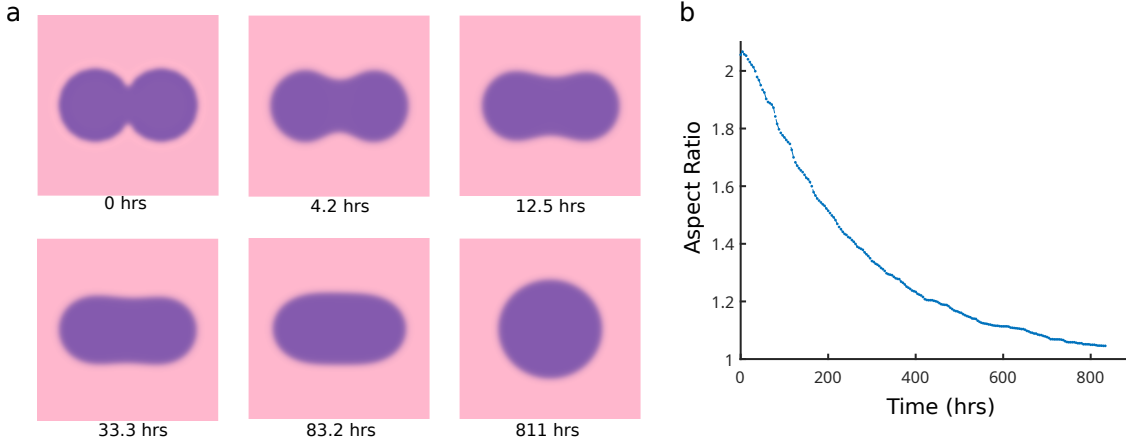


Figure 8.2: Estimating Surface Tension: (a) Time snapshots from simulations showing the merging of two cancer droplets due to surface tension. In all panels, cancer cells are shown in magenta and the ECM is shown in light pink. (b) Aspect ratio of the droplets decreases as a function of time, as the drop becomes more and more circular.

$\Delta t = 30\text{ s}$, and $\Sigma = 4\text{ kPa}$ respectively. We use the elastic modulus $E = 0.1 = 400\text{ Pa}$, and activities in the range $\zeta \in [0.007, 0.2] = [28\text{ Pa}, 800\text{ Pa}]$. The crucial non-dimensional parameter here is the ratio of the active driving stress and the opposing elastic stress, varied here between $\zeta/E \in [0.07, 2]$.

We further use $\rho = 40$, $\nu = 1$, $\eta = 40$, $A_\varphi = K_\varphi = 1$, and $\Gamma_\varphi = 0.33$ for the remaining fluid parameters. The nematic parameters for the cancer clusters are chosen to be $C^c = 0.1$, $K_{LC}^c = 0.1$, $S_{c,0} = 1$, $\lambda^c = 0.1$, and $\Gamma = 0.33$. These are typical parameters for a nematic liquid crystal in an ordered phase, with characteristic lengthscales of a few lattice widths ($\sim 10\text{ }\mu\text{m}$). For the ECM, we use $C^e = 0.02$, $K_{LC}^e = 0.01$, $S_{e,0} = 0$, and $\lambda^e = 0.1$, with the weak free energy chosen to isolate the effect of active flows on the ECM. All simulations are run with the above-mentioned parameters unless stated otherwise.

With these chosen parameter values, the typical active velocities are $\approx 6 \times 10^{-3}$ (corresponding to $3.6\text{ }\mu\text{m hr}^{-1}$). The second parameter to fit is the surface tension. This is numerically estimated by initializing two droplets adjacent to each other, and measuring the time when the notch between the two disappear. For initially touching droplets of radii 20 ($= 100\text{ }\mu\text{m}$), they ‘merge’ in $\approx 6 \times 10^3$ LB timesteps (corresponding to $\sim 50\text{ hrs}$, see Fig. 8.2), which is comparable to the results reported from cancer spheroid experiments ($\sim 36\text{ hrs}$) [197].

We ran simulations in a periodic box of size 400×400 for 400,000 timesteps, with snapshots taken every 2000 timesteps. For simulations of droplet breaking, we initialized the system with a circular droplet with radius 100, covering $\sim 20\%$ of the simulation box. For simulations of droplet coarsening, we initialized the system with a uniform value of $\phi = 0.25$ everywhere, resulting in many small droplets of $\phi = 1$ covering 25% of the simulation domain. For the simulation results in Fig. 4, we ran simulations starting from a circular cluster of radius $R = 40$ in a periodic box of size 200×200 for 84,000 timesteps, with snapshots taken every 2000 timesteps. In all cases, we initialized the nematic director field at random, and simulated for 100 pre-initialization steps without activity.

8.3 Shapes and Sizes of Cancer Clusters

In this section, we compare simulation results of cancer cluster sizes and shapes to the ones obtained from histological samples. To do this, we study the more realistic case of multiple cancer clusters with different activities confined by viscoelastic ECM.

8.3.1 Dynamical Steady States

A large cancer cluster with high activity breaks up over time into many small active clusters (See Fig. 8.1b). Active forces make these clusters highly dynamic, making them constantly moving around and forming active protrusions. Surface tension forces split up these protrusions to form smaller active clusters, and lead to the coalescence of touching clusters to form larger aggregates.

This results in a steady distribution of shapes and sizes of cancer clusters, known as a dynamical steady state. Interestingly, at long enough times, the details of the dynamical steady state do not retain any memory of the initial condition, and only depend on the activity of the active droplets. The same steady state can also be achieved by running simulations with many small active droplets, which coarsen over time until they arrive at a dynamical steady state [101].

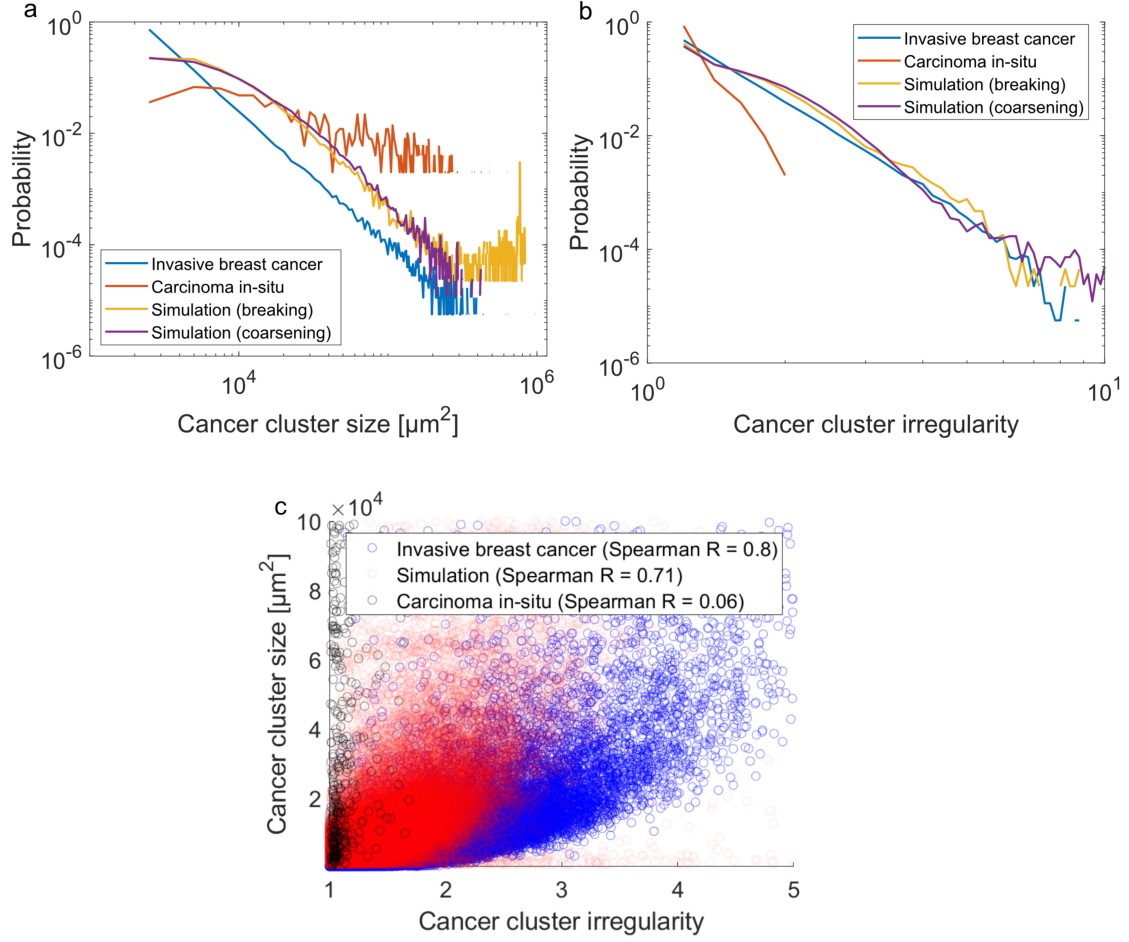


Figure 8.3: Comparing simulation results to histological data: (a) Distribution of tumour sizes in histological samples (blue) compared with simulation results (yellow and purple) from different initial conditions. Larger droplets follow the same power law distribution. Red curve shows non-malignant in-situ breast tumours (DCIS). (b) Distribution of tumour shapes (characterized by irregularity) in histological samples (blue) and simulation results (yellow and purple) agree very well. (c) Shapes and sizes of cancer tumours are strongly correlated in both histological (blue) and simulation (red) data. Non-malignant DCIS tumours do not show this behaviour.

8.3.2 Comparing Shapes and Sizes

The simulation data is obtained by averaging dynamical steady-states of multiple droplets across a range of activities. The details of activity averaging are discussed in Sec. 8.3.3.

In order to compare simulation data to experimental data for the cross-section sizes, we need to be aware that we are comparing two-dimensional cross-sections of three-dimensional sizes in the histological samples to real two-dimensional systems

in the simulations. We discuss this further in Sec. 8.3.4 .

The distributions of sizes observed from the simulations follow a power law, spanning more than a decade and covering a large range of droplet sizes. A similar power law, with comparable exponents, is observed from histological samples. These results are compared in Fig. 8.3a.

We find that our simulation size distribution deviates from the observed power law result for small cancer clusters. We attribute this to the resolution of the cancer-ECM interface in our simulations, which is smoothed out over roughly 3 lattice points. Only accounting for clusters with a radius ≥ 5 LB units removes this discrepancy and gives a very good fit. It is also easy to numerically simulate higher surface tensions, leading to interfaces smoothed over more lattice sites. The higher surface tension increases the mismatch between the simulations and the power law, and a larger cluster cutoff size is required after which the power law emerges, consistent with expectations.

Cancer clusters in histological samples tend to form irregular tortuous shapes. We quantify this feature by defining a dimensionless shape index for the cancer clusters called the irregularity,

$$\text{Irregularity} = L/\sqrt{4\pi A}, \quad (8.6)$$

where L is the perimeter of the cluster, and A is the cross-sectional area. This quantity is 1 for circular droplets and increases to higher values as the shape of the cluster gets more tortuous.

We compute the shape factor from histological samples and simulation data, plotting them together in Fig 8.3b. We find that the simulation data agrees well with the clinical data.

Finally, sizes and shapes are strongly linearly correlated in droplet simulations, quantified by calculating the Pearson correlation coefficient R^2 between the two datasets. We explain this by considering that the roughness of the boundary is driven by fingering instabilities at the boundary [111], the number of which scales with the radius of the droplets. Therefore, larger droplets tend more protrusions,

leading to more irregular tortuous shapes in simulations. This positive correlation is also seen in clinical samples. The results from simulation data are plotted over the histological data are compared in Fig. 8.3c. The simulation and clinical data have significant overlap, and both have strong correlations between shape and size. However the clinical data also shows a number of clusters with higher irregularity at the same size. This would be consistent with some highly active cancer clusters in the histological samples highly fluidizing the nearby ECM, to yield lower values of elasticity and correspondingly more irregular clusters.

8.3.3 Activity Averaging

In clinical samples, not all cell colonies have the same motility, but instead the colonies have a spread of motility [196, 197]. Thus, to compare simulation results to patient-averaged clinical data, we should average over a distribution of activities instead. Therefore, we average simulation results over a distribution of activity to compare to the histological data.

In Fig. 8.4a, we find that changing the exact form of the activity distribution does not make a qualitative difference in the distribution of size data, as long as the sampling includes clusters with intermediate activity. Different sampling schemes have different area fractions of cancer clusters with a given activity. These are labelled by different colours, and the relative weights for different activity values are shown in Fig. 8.4b. Systems with higher activity have a significantly higher number of cancer clusters, which are smaller in size (Fig. 8.4c). These form the large majority of the observed clusters, dominating the statistics of the overall distribution. In the results presented in this chapter, we have used the uniform scheme ('Scheme 1') to capture a broad spread of activity in cancer clusters.

We also find after averaging over activity, our simulation results are robust to changing the ECM elasticity and the concentration fraction of cancer clusters. This may be because activity is a critical parameter in all these simulations, and averaging over activity scans over the entire range of shapes and sizes observed with different activity numbers.

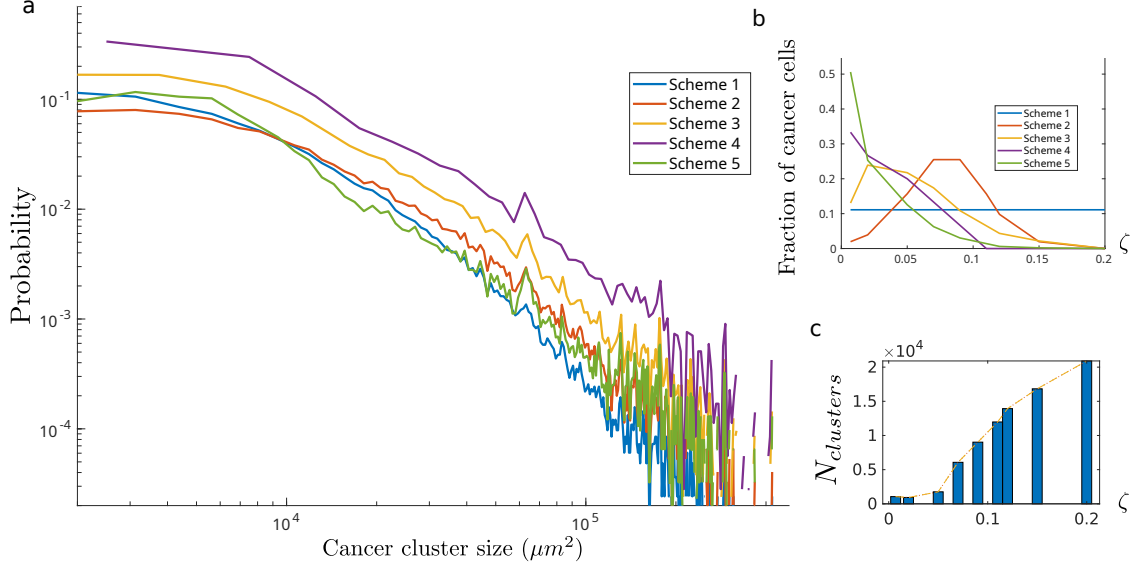


Figure 8.4: (a) Observed size distributions of cancer clusters for five different schemes of activity averaging, represented by different colours, all showing a similar power-law distribution. (b) Different activity averaging schemes, showing relative fractions of different activities in the system. (c) Bar plot showing the number of cancer clusters increases with activity, for a fixed area fraction of cancer cells.

It is worth pointing out with a concrete example that activity averaging can yield a power law of observed cluster sizes. This is because while simulations with different activity might have different intrinsic lengthscales, averaging over activity can wash out this preferred lengthscale resulting in a power law distribution which is scale invariant. Below, we consider an example which shows that activity averaging can give rise to such a power law.

Let the distribution function of activities of cancer clusters in the sample be given by $p(\zeta)$ i.e. the probability of a randomly chosen cluster having activity in the range $[\zeta, \zeta + d\zeta]$ is $p(\zeta)d\zeta$. We find from simulations that a large and active cancer droplet breaks up into many droplets in a dynamical steady state. Let the number of smaller cancer clusters, which depends on the activity ζ , be given by the function $n(\zeta)$. When the confinement is viscous, we find numerically that the resultant smaller droplets follow an exponential size distribution, given by $f_\zeta(A) = k(\zeta) e^{-k(\zeta)A}$. Here, A is the area of the resultant droplets, and the preferred scale $k(\zeta)$ is also a function of the activity.

Given this distribution of activities, the total number of expected clusters of size A is given by

$$N(A) = \int_{\zeta} f_{\zeta}(A) n(\zeta) p(\zeta) d\zeta = \int_{\zeta} k(\zeta) e^{-k(\zeta)A} n(\zeta) p(\zeta) d\zeta \quad (8.7)$$

and the probability distribution of observing a cluster of size A is obtained by simply normalizing,

$$\rho(A) = \frac{N(A)}{\int_{A_0} N(A_0) dA_0}. \quad (8.8)$$

To proceed further from this point, we need to make assumptions about the missing distribution functions in Eq.(8.7). One simplifying assumption is to assume that the activity (motility) is uniformly distributed within a range of values:

$$p(\zeta) = \begin{cases} \frac{1}{\zeta_{max} - \zeta_{min}} & \zeta \in [\zeta_{min}, \zeta_{max}], \\ 0 & \text{otherwise.} \end{cases} \quad (8.9)$$

Analytically, we further assume that $k(\zeta) = \alpha\zeta$ and $n(\zeta) = \beta\zeta$. Thus,

$$N(A) = \int_{\zeta_{min}}^{\zeta_{max}} \frac{\alpha\beta}{\zeta_{max} - \zeta_{min}} \zeta^2 e^{-\zeta\alpha A} d\zeta \approx \frac{\beta \Gamma(3)}{\alpha^2 (\zeta_{max} - \zeta_{min})} \frac{1}{A^3}$$

which is proportional to A^{-3} . To arrive at this result, we have changed variables to $t = \zeta\alpha A$, and then approximated the integral limits to run from 0 to ∞ , due to the wide range in A spanning over two decades.

We expect to find different power laws for a range of different functions $k(\zeta)$, $n(\zeta)$ and $p(\zeta)$, since the argument revolves around non-dimensionalizing the integral, and approximating the upper and lower limits of t exploiting the fact that the exponential drops off quickly over the wide range of A . While this calculation demonstrates that activity averaging can result in power-law distributions, the exponent predicted by this calculation is -3 . The exponent obtained from histological samples is -2.4 , which is the same as the exponent obtained from numerically averaging simulation results for different activities, as shown in Fig. 8.4.

8.3.4 Mapping three-dimensional snapshots to two dimensions

The clinical images are two-dimensional projections of three-dimensional cancer clusters. However, the simulations consider two-dimensional clusters. The study of mapping from one to another is called stereology [200]. An exact mapping can be derived for collections of perfect spheres [201], and an analogous mapping can be derived for ellipsoids oriented isotropically [201, 202]. For irregular shapes, studies show a strong correlation between the 2d observed size distributions and the real 3d volume distributions [203].

As a specific example, we discuss the case of spherical clusters following the analysis in Ref. [201], and show below that the observed size distribution remains unchanged when looking at randomly chosen cross-sections. Consider a sphere of radius R and central cross-section area $A_0 = \pi R^2$. We observe a cross-section of the sphere by cutting through a plane at distance d from the centre of a sphere of radius R . The observed cross-sectional area is then $A = \pi(R^2 - d^2)$, and hence $d = \sqrt{\frac{A_0 - A}{\pi}}$.

We now assume the probability distribution of the d to be uniformly distributed across the radius of the sphere,

$$p_d(d) = 1/R, \quad 0 < d < R. \quad (8.10)$$

Changing variables to A , we find

$$p(A|A_0) = \frac{1}{2} \frac{1}{\sqrt{A_0(A_0 - A)}}, \quad A < A_0. \quad (8.11)$$

This is the probability of observing a cross-sectional area of A , for a sphere with a central cross-sectional area A_0 . p is a probability density function and is allowed to diverge at a point A_0 , as long as the total probability density is normalizable.

Finally, the observed distribution of A , for a distribution of sphere cross-sections $p(A_0)$, is given by

$$p(A) = \int_A^\infty p(A|A_0)p(A_0) dA_0. \quad (8.12)$$

If $p(A_0)$ is distributed like a power law, then Eq. 8.12 implies $p(A)$ is also distributed as a power law with the same exponent.

8.4 Fingerprints of active driving in the ECM

Although our simulated cluster sizes and shapes are a good match with the distributions obtained from histology, we do not have any clinical data about the dynamics of such systems. Instead, we compare other predictions made by our model with the histological samples. In this section, we look at the effect of active flows on the orientation field of the ECM in the vicinity of an active cluster. To do this, we simulate a single cluster with $R = 40 LB$ units, and set $K_{LC} = 0.01$ to have large active droplets which are not broken apart by active fingering instabilities.

The ECM orientation field was extracted from the eosin intensity data in histological samples by our collaborators, using the methods developed in Ref. [204]. This data was then digitally analyzed to analyze nematic topological defects and anchoring angles near malignant clusters.

8.4.1 Topological defects near cancer clusters

The active flows generated by cancer clusters also affect the ECM through the continuity of stress at the boundary. These drive flows in the ECM, which acts like a passive driven material. The vorticity and strain-rate tensor of the ECM flow field affects the orientation field of the ECM. These flows can give rise to topological defects in the ECM orientation field around cancer clusters.

The defects are formed mostly close to the cluster, where the flows are largest in magnitude. The defect density drops exponentially with distance from the cancer cluster. This distance increases upon increasing the activity of a cluster and decreases upon increasing the elasticity of the ECM. Our results show good quantitative agreement with the defect density measured close to cancer clusters in histological samples, as shown in Fig. 8.5.

While we were unable to find an analytical lengthscale for this defect distribution, it is possible to make qualitative arguments about the lengthscale. Since the ECM is a driven material, the magnitude of flow decays with distance from the cancer cluster. Larger flow magnitudes result in larger perturbations in the ECM orientation field, leading to more defects. Thus, higher activities give rise to more defects, and

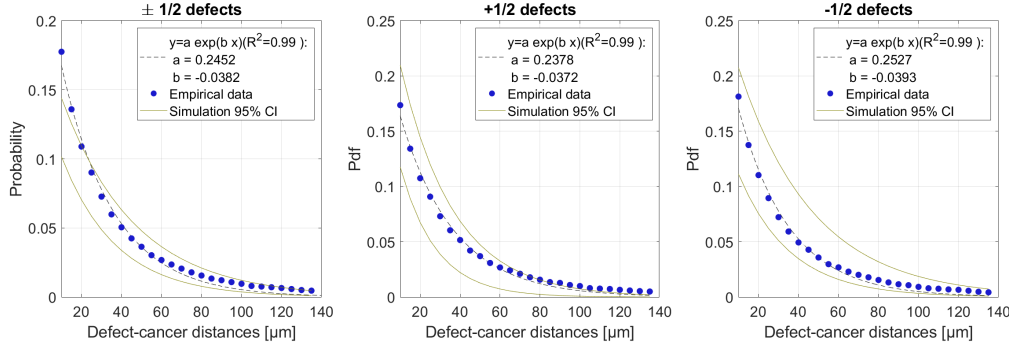


Figure 8.5: Topological Defects in ECM: Topological defect densities decay exponentially with distance from the cancer tumour for both simulation and histological data. Data from histological samples is marked in blue. The green lines show the 95% CI of the probability distributions of the defect-cancer distances, for simulations of a single droplet in an elastic ECM.

stronger flows result in more defects further away from the clusters. On the other hand, higher ECM elasticity offers greater resistance to flow, which decays quickly away from the driving source. This leads to fewer defects closer to the cancer cluster. Our simulation results agree with these qualitative expectations. The difficulty in making these numbers more precise is that the argument requires that the system fluctuates dynamically, as a steadily driven Kelvin-Voigt material has no flows in the infinite time limit.

8.4.2 ECM anchoring at cluster boundaries

Individual cancer clusters change their shape due to active flows. Some regions push out the ECM to form protrusions, and in other regions create inwards flows pulling in the ECM. This drives flows in the ECM which anchor the ECM orientation field at the boundary of the cancer cluster.

We define the ECM anchoring angle θ_a as the angle made by the ECM orientation field to the tangent vector of the cancer cluster. Parallel anchoring corresponds to $\theta_a = 0^\circ$, and perpendicular anchoring corresponds to $\theta_a = 90^\circ$.

The flow-alignment term tends to orient the ECM parallel to the cluster interface for flows pushing radially outwards, and perpendicular to the interface for flows pulling radially inwards. Consider, for example, a radial velocity field inside the ECM that decays with distance from the origin, $v_r = k/r$. The strain-rate tensor is

given by $\mathbf{E} = -\frac{k}{r^2}\hat{\mathbf{r}} \otimes \hat{\mathbf{r}} + \frac{k}{r^2}\hat{\boldsymbol{\theta}} \otimes \hat{\boldsymbol{\theta}}$. The largest eigenvalue of $\mathbf{E}$ is radial if $k < 0$ (pulling), and tangential if $k > 0$ (pushing). A similar argument holds for all radial flow profiles which decay into the ECM as $v_r \sim r^{-\alpha}$, where $\alpha \geq 0$, but breaks down when $\alpha < 0$. The orientation field aligns with the largest eigenvalue of $\mathbf{E}$, aligning the ECM at the cluster boundaries.

We compare the distribution of θ_a obtained from simulations with data obtained from clinical samples. Both show similar quantitative properties (see Fig. 8.6a), with a median value $\theta_a \approx 33^\circ$, and a distribution skewed towards parallel anchoring. Moreover, the orientation field at the boundary shows alternating regions of parallel and perpendicular anchoring for both numerical simulations and clinical data (see Fig. 8.6b and Fig. 8.6c respectively), reminiscent of active fingering at droplet boundaries [111].

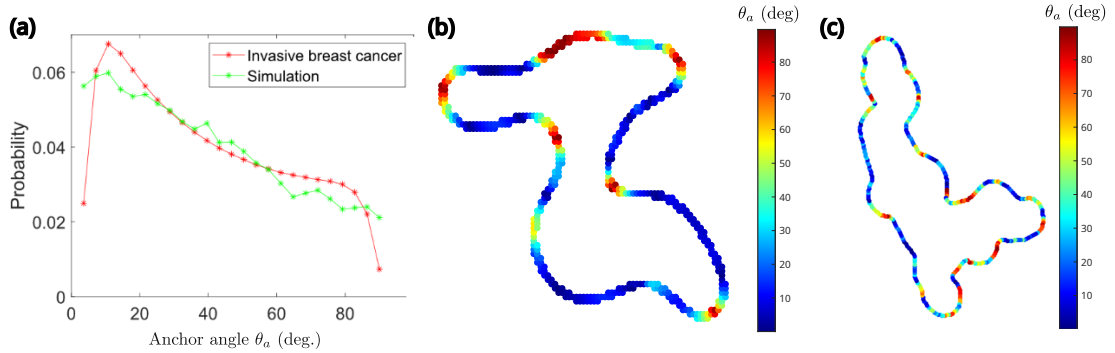


Figure 8.6: ECM Anchoring at Cancer Tumour Boundaries: (a) Observed distribution function of anchoring angle θ_a . The clinical data (red) is averaged over all clinical samples, while the simulation data (green) shows the time-averaged anchoring angles for a single cancer tumour. The median value for both distributions is 33° . (b) Snapshot of a simulated cancer cluster, with colouring showing anchoring angle θ_a at the cluster boundary. (c) Snapshot taken from a clinical sample, with colouring showing anchoring angle θ_a at the cancer cluster boundary. Both (b) and (c) show qualitatively similar properties, with curved regions having $\theta_a \approx 90^\circ$, and long straight regions with $\theta_a \approx 0^\circ$.

8.5 Motility or Growth

At first glance, it might be surprising that the active nematic model fits the observed data, because it does not account for the fact that cancer clusters are also growing.

However, the active flows that determine the dynamical steady state distribution are dominated by the effect of motility.

The typical velocity scale for motility in a cancer cluster is $v_{\text{motility}} \sim 3 \mu\text{m hr}^{-1}$. We can estimate the typical proliferation velocities in two ways - by looking at mitosis markers in cells, which indicate the fraction of cells undergoing cell division, and by looking at the overall growth over time of macroscopic tumours. Both methods yield an estimate of around $0.04 - 0.10 \mu\text{m hr}^{-1}$, which is orders of magnitude smaller than flows due to motility. This expectation is borne out by the observation that droplet sizes are independent of the proliferation rate of the cancer tumour. We see an inverse correlation of droplet size with the magnitude of the activity in simulations, as increasing activity values make large clusters more unstable.

Although the flows due to growth are too slow to affect the dynamical steady state of the system, they do change the overall area fraction of the system at a slower rate than the active dynamics. However, we have checked that changing the area fraction does not quantitatively affect the underlying distributions for the shapes and sizes of cancer clusters.

8.6 Summary and Discussion

In this chapter, we modelled malignant breast cancer tumours inside a viscoelastic extracellular matrix (ECM) as a confined active nematic. We showed that a minimal model of active nematics confined by a Kelvin-Voigt viscoelastic material results in a dynamical steady state which accurately reproduces the sizes and shapes distributions of cancer tumours observed in clinical samples. We then investigated the effects of activity on the surrounding ECM, showing that active flows create topological defects and align ECM fibres at the cancer-ECM boundary. We conclude by arguing that motility, instead of growth, is the ‘activity’ relevant to understanding the dynamics of this system. Our results highlight the potential of using active matter physics to study the physics of cancer migration and metastasis.

Take-home message:

Modelling cancer tumours inside the ECM as an active droplet confined inside a passive viscoelastic medium reproduces features observed in clinical samples.

“For once you have tasted flight you will walk the earth with your eyes turned skyward; for there you have been, and there you long to return.”

— “I, Leonardo da Vinci” (1965 biography)

9

Discussion

In this thesis, we have used active nematics to explore various consequences of active flows in living systems. We looked at the effects of activity on patterning, showing how active flows can both support and hinder patterning. We also looked at phase separation in active nematics, and how confinement can lead to regular phase-separated patterning. Finally, we considered the mechanical coupling of an active system with external viscoelastic confinement, and applied this model to study breast cancer tumours embedded inside an extracellular matrix. We summarise the main findings of each chapter in Sec. 9.1. Finally, we conclude by highlighting key open questions about the effects of cellular heterogeneity and the interplay between activity and viscoelasticity in active models of morphogenesis and cancer metastasis in Sec. 9.2.

9.1 Summary

In Chapter 3, we used a two-fluid model with compressible components to study homogeneous mixtures of an active nematic and a passive isotropic species. We found that this system spontaneously phase separates into domains with high active concentration even if it is energetically unfavourable to do so. Active nematics align with the concentration gradient at the interface between the phases, called ‘active

anchoring'. Due to active anchoring, normal active forces at the interface tend to magnify the concentration difference between the two phases. We found that the resultant micro-phases form at the active lengthscale, and investigated how the magnitude of phase separation and the sizes of the resultant droplets vary with the thermodynamic free energy, viscous drag between fluids, and surface tension between the fluids. Finally, we looked at the effect of interfacial curvature on phase separation, showing that interfaces with negative curvature enhance the phase separation mechanism, while interfaces with positive curvature have weaker phase separation.

In Chapter 4, we extended the results of the previous chapter to study mixtures where both species are active. Using numerical simulations and analytical arguments, we showed that the orientation fields of two active species develop an effective alignment or anti-alignment interaction due to the coupling by viscous drag. We found that the alignment between director fields plays an important role in the phase separation of active-active mixtures. In the absence of imposed director alignment, the magnitude of phase separation is greatest when one species is active and the other species is passive. However, with elastic alignment between the two species the magnitude of phase separation is greatest when one of the species is extensile and the other contractile. We then investigated how the magnitude of phase separation and the morphology of the resultant phases can be controlled by changing the relative activities, relative concentration fractions, or by adding frictional forces with an underlying substrate.

In Chapter 5, we showed how mixtures of active nematic and passive isotropic species under confinement spontaneously phase separate, sorting the active component into specific regions. We showed that the active component sorts itself in the centre of a channel with laminar flows, and near the edge for a spontaneously rotating active droplet in viscous confinement. We further found that even active turbulent states preferentially sort the active component to the wall, under appropriate nematic anchoring. Our results show how active forces can give rise to phase ordering in active nematic mixtures.

In Chapter 6, we looked at the interplay of chemical signalling with active nematic cell layers. We found that active flows can affect the morphology and strength of Turing patterns, with the stripes bending and breaking until they ultimately dissolve above a critical activity. We showed that the magnitude of Turing patterns can be quantified by a single Karlovitz number, characterizing the relative timescales of active advection and the reaction flux. We then investigated the effects of coupling the nematic activity with the concentration of morphogen. We found that when Turing stripes with different morphogens have extensile and contractile activity respectively, the system self-organizes into a configuration with shearing flows inside each stripe, with characteristic slipping instabilities. Our results show that mechano-chemical coupling in active systems can lead to novel flow states.

In Chapter 7, we turned our attention to mechanical coupling between active nematics and their confinement. We numerically studied an active nematic in a channel confined by viscoelastic walls. We found that this system exhibits periodic flow reversals over a range of activity and elasticity, arising from the feedback between active flows in the channel and elastic restoring forces from the surroundings. Our results are an example of how mechanical feedback can lead to new spatiotemporal flow states in active matter.

Finally, in chapter 8, we modelled malignant breast cancer tumours inside a viscoelastic extracellular matrix (ECM) as a confined active nematic. We found that active droplets continually split and merge, giving rise to a dynamical steady state which accurately reproduces the sizes and shapes of cancer tumours obtained from patient data. We then investigated the effects of active flows on the confining ECM - in particular, looking at the density of topological defects and the orientation of ECM fibres near cancer clusters. Our results matched closely with patient data. We finally concluded this chapter by showing that motility, instead of growth, dominates the active dynamics of these tumours. Our results show that active matter physics may lead to new insights on tumour dynamics and cancer metastasis.

9.2 Outlook

Active hydrodynamic models are increasingly being used to describe biological tissues and incorporate key biological features such as cell motility, division and death. In this thesis, we have touched upon some of the implications of modelling biological tissues as active nematics. However, there are still many open questions which are yet to be answered. Some important examples include:

How does cellular heterogeneity in cell aggregates affect dynamics?

Cellular aggregates are usually not homogeneous but consist of many distinct sub-populations of cells with different phenotypes. These sub-populations can have different mechanical properties, chemical properties and cellular responses [205, 206]. Moreover, the populations inside growing cell aggregates can change over time [207] as cells differentiate, migrate and divide. While we have made some progress on this question by studying mixtures of two active cell types, it would be interesting to extend these ideas further to study the consequences of cellular heterogeneity in active systems.

Can spatiotemporal control of activity and elasticity guide tissue flow in morphology?

Recent work has shown that active matter in three dimensions can replicate key features of morphogenesis like arm-like protrusions [32] and invagination [208] in active nematic droplets. Other recent work has shown that elasticity plays a key role in morphogenetic flows, with cells undergoing a fluid-to-solid jamming transition [209].

It would be interesting how activity and elasticity interact with each other to drive active flows in morphogenesis. One interesting question to ask is if (and how) spatiotemporal activity $\zeta(\mathbf{x}, t)$ and local elasticity $E(\mathbf{x}, t)$ patterns in an active droplet can be tuned to achieve the flow patterns and final shapes observed in morphogenesis.

Can theories of active matter in viscoelastic confinement help us understand cancer progression?

In this thesis, we have shown that active matter in viscoelastic confinement can be used as a model to study breast cancer tumours in the ECM. The cancer cluster applies active forces on the ECM, modifying its structure and generating topological defects. The ECM, in turn, applies forces on the cancer cluster and suppresses active instabilities, thus changing the dynamics of the cancer cluster. This feedback between active matter and its confinement is poorly understood.

Another promising avenue for future research is to study the effects of motility and growth in other phases of breast cancer progression. For instance, when healthy breast duct epithelia become cancerous [210], the cells start proliferating uncontrollably, rapidly filling in the lumen inside the duct with epithelial cells to form a breast cancer tumour. It would be interesting to try and study this process using an active matter framework.

Appendices

“ We must use sometimes the one theory and sometimes the other, while at times we may use either.”

— Albert Einstein, on wave-particle duality



The hybrid Lattice-Boltzmann method

In this thesis, we have numerically solved the Navier-Stokes equations 2.2 and mass continuity equation 2.1 for the flow field simultaneously using a Lattice-Boltzmann algorithm. For all the other fields in the system, we have used a finite difference method to solve the equations of motion. This approach, pioneered by Marenduzzo et al. [33, 211], is called the hybrid Lattice-Boltzmann method.

A.1 From Navier Stokes to Lattice Boltzmann

The Lattice-Boltzmann (LB) method is a numerical way to solve the Navier-Stokes equations using particle-based methods. Instead of solving the equations of motion directly, this method instead solves a discretized version of the Boltzmann equation. The Boltzmann equation describes how momentum distributions of particles $f_{\mathbf{k}}(\mathbf{r})$ evolve over time due to advection and collisions with other particles. Here, $f_{\mathbf{k}}(\mathbf{r})$ is the number of particles of momentum $\mathbf{k}$ at location $\mathbf{r}$. The Navier-Stokes equations arise as a special case of the Boltzmann equation under the following assumptions:

1. The distribution function is close to a local equilibrium function $f_{\mathbf{k}}^0$. Thus,

$$f_{\mathbf{k}}(\mathbf{r}) = f_{\mathbf{k}}^0 + \delta f_{\mathbf{k}}^1(\mathbf{r}) \tag{A.1}$$

2. The collision term relaxes the distributions to the equilibrium function over a relaxation time τ . Thus,

$$\mathcal{I}_k[\{f_i\}] = -\frac{f_k(\mathbf{r}) - f_k^0}{\tau} \quad (\text{A.2})$$

These two assumptions yield the Navier-Stokes equations, after using the Chapman-Enskog expansion [212]. The Lattice-Boltzmann (LB) method is computationally efficient, easily parallelized, and is a second-order accurate solver for the weakly-compressible Navier-Stokes equation [52].

A.2 The Lattice-Boltzmann equation

In the LB algorithm, the distribution function $f_k(\mathbf{r})$ is only defined over discrete lattice sites $\mathbf{r}$ with spacing Δx . Moreover, momentum space is discretized into a fixed set of direction vectors $\mathbf{e}_i$, where index i labels specific lattice direction. In 2D, we use the ‘D2Q9’ velocity vector set $\mathbf{e}_i = (0, 0)$, $(\pm 1, 0)$, $(0, \pm 1)$, and $(\pm 1, \pm 1)$, where i runs from 0 to 8 (see Fig. A.1). Each direction also has an associated weight w_i (see Fig. A.1). Thus, we arrive at the distribution function $f_i(\mathbf{x}, t)$, describing the number of particles at lattice site $\mathbf{x}$ at time t moving with velocity $\mathbf{e}_i$.

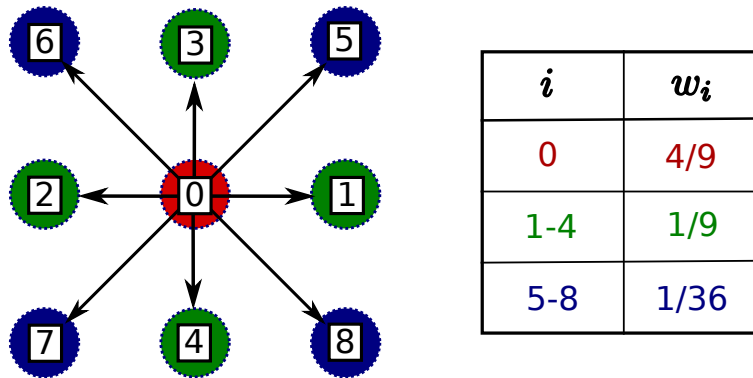


Figure A.1: Lattice geometry and weights used for the D2Q9 lattice Boltzmann model. The zero node (red) corresponds to particles with no momentum.

The fluid density ρ is related to the distribution function by

$$\rho(\mathbf{x}, t) = \sum_i f_i(\mathbf{x}, t), \quad (\text{A.3})$$

and the fluid momentum is given by

$$\rho \mathbf{u} = \sum_i f_i \mathbf{e}_i, \quad (\text{A.4})$$

where $\mathbf{u}$ is the flow field.

The time evolution of the distribution function $f_i(\mathbf{x}, t)$ has two steps:

1. Collision step: Update the local distribution functions due to collisions

$$\tilde{f}_i(\mathbf{x}, t) = f_i(\mathbf{x}, t) + \Delta t C_f(\mathbf{x}, t). \quad (\text{A.5})$$

2. Streaming step: Move each particle to the next lattice site in the direction of motion

$$f_i(\mathbf{x} + \mathbf{e}_i \Delta t, t + \Delta t) = \tilde{f}_i(\mathbf{x}, t). \quad (\text{A.6})$$

In Eq.(A.5), C_f is a collision operator which captures the local relaxation to equilibrium, but also contains an additional source term $S_i(\mathbf{x}, t)$ to model body forces,

$$C_f(\mathbf{x}, t) = -\frac{1}{\tau} (f_i - f_i^0) + S_i(\mathbf{x}, t). \quad (\text{A.7})$$

There are many forms of collision operators and different schemes to model body forces as source terms. We have used the Bhatnagar-Gross-Krook (BGK) collision operator (Eq. A.7), and a Guo forcing scheme [186]. Moreover, the equilibrium distributions f_i^0 must satisfy

$$\sum_i f_i^0 = \rho, \quad \sum_i \mathbf{e}_i f_i^0 = \rho \mathbf{u}, \quad \sum_i \mathbf{e}_i \mathbf{e}_i f_i^0 = -\boldsymbol{\sigma} + \rho \mathbf{u} \mathbf{u}, \quad (\text{A.8})$$

where $\boldsymbol{\sigma}$ describes the symmetric terms of the stress tensor in thermodynamic equilibrium. We use the equilibrium function

$$f_i^0 = w_i \rho \left(1 + \frac{\mathbf{u} \cdot \mathbf{c}_i}{c_s^2} + \frac{(\mathbf{u} \cdot \mathbf{c}_i)^2}{c_s^4} - \frac{\mathbf{u} \cdot \mathbf{u}}{2c_s^2} \right) \quad (\text{A.9})$$

where w_i are the ‘D2Q9’ weights and c_s is the speed of sound in LB units [52].

A.2.1 Boundary Conditions

Rigid walls with free-slip or no-slip boundary conditions are treated in Lattice-Boltzmann by adding an additional layer of ghost nodes inside the boundary, where we are free to tune the distribution functions f_i . By a judicious choice of these functions, the velocity field at a virtual boundary node in between the last fluid node and the ghost node can be controlled as desired. We implemented this method, called the bounce-back method, to study free slip and no-slip boundary conditions for the velocity field at walls. We refer the reader to Chapter 5 in [52] for a more detailed explanation of this method.

A.3 Lattice-Boltzmann for the Two-Fluid model

We follow the method prescribed by Refs.[97, 98] to simulate a two-fluid model, while keeping track of additional forcing terms due to substrate friction and active forcing, as discussed in Sec. 3.2.

The combined fluid is incompressible and is solved using an LB algorithm. However, the equilibrium function (see Eq. A.8) needs to be modified to account for the surface tension term in σ^{thermo} (see Eq.(3.10)). This new equilibrium function is given by [97]

$$f_i^0(\rho, \mathbf{u}) = \rho w_i \left[1 + 3(\mathbf{e}_i \cdot \mathbf{u}) - \frac{3}{2} \|\mathbf{u}\|^2 + \frac{9}{2} (\mathbf{e}_i \cdot \mathbf{u})^2 \right] + \frac{9}{2} w_i \kappa \left[(\mathbf{e}_i \cdot \nabla \phi)^2 - \frac{1}{2} \|\mathbf{e}_i\|^2 \|\nabla \phi\|^2 \right] \quad (\text{A.10})$$

However, in addition, we now have to keep track of a compressible component fluid. We achieve this by using two additional distribution functions g_i and $\tilde{g}_i$, to keep track of the density and momentum of the component fluid respectively [97] through the equations

$$\rho^0(\mathbf{x}, t + \Delta t) = \sum_i \tilde{g}_i(\mathbf{x} - \mathbf{e}_i \Delta t, t) \quad (\text{A.11})$$

$$\rho(\mathbf{x}, t + \Delta t) \mathbf{u}^0(\mathbf{x}, t + \Delta t) = \sum_i \mathbf{e}_i \frac{g_i(\mathbf{x} - \mathbf{e}_i \Delta t, t)}{\phi(\mathbf{x} - \mathbf{e}_i \Delta t, t)} + \mathbf{G} \Delta t \quad (\text{A.12})$$

The streaming step for the momentum equation (Eq. (A.12)) is modified to account for compressibility. The distribution functions g and $\tilde{g}$ are given by [97]

$$g_i(\boldsymbol{x}, t) = f_i^0(\rho^0, \mathbf{u}^0), \quad \tilde{g}_i(\boldsymbol{x}, t) = f_i^0(\rho^0, \mathbf{u}^0 + \frac{1}{6} \ln \rho^0 \Delta t) \quad . \quad (\text{A.13})$$

*“Two roads diverged in a wood, and I —
I took the one less traveled by,
And that has made all the difference.”*

— Robert Frost (The Road Not Taken)

B

Overview of Models

In this appendix, we summarize the different models used throughout this thesis, highlighting the different models and configurations we have used in each chapter.

B.1 Models at a Glance

B.1.1 Active Nematics

Throughout this thesis, we use active nematics as the model for studying living matter. This model is introduced in Chapter 2, and describes the coupled dynamics of the nematic tensor, $\mathbf{Q}$, and the hydrodynamic velocity field $\mathbf{u}$. The dynamics of the nematic tensor, $\mathbf{Q}$, is described by the Beris-Edwards equation (Eq.(2.12)), while the velocity field evolves according to the Navier-Stokes equations with additional forcing terms (Eqs.(2.1)-(2.2)). Active nematic forces are modelled by the active stress term (Eq.(2.18)), where ζ is the activity coefficient describing the strength of the individual dipoles. These equations are solved using the methods described in Sec. 2.7.

B.1.2 Two-Fluid model

In Chapters 3–5, we use a two-fluid model to study a heterogeneous mixture of two different species which can flow past each other, described in Sec. 3.2. Each species is labelled by an index $i \in \{1, 2\}$, and has an associated density field ρ^i ,

velocity field $\mathbf{u}^i$, and nematic tensor $\mathbf{Q}^i$. The local concentration fraction of the first component fluid is given by $\phi = \frac{\rho^1}{\rho^1 + \rho^2}$.

Each component fluid satisfies mass conservation (Eq.(3.1)) and momentum balance (Eq.(3.2)). The two fluids are coupled by viscous drag (the magnitude of which is given by γ), a thermodynamic free energy, and the constraint of combined incompressibility ($\rho^1 + \rho^2 = \text{const}$). Each nematic field satisfies a Beris-Edwards equation, modified to account for the compressibility of the component fluid (Eq.(3.19)). The whole system is simulated using a modified hybrid Lattice-Boltzmann scheme with three distribution functions, described in Sec. 3.2.

B.1.3 Reaction-Diffusion models

In Chapter 6, we use two chemical fields to study the interaction of Turing patterns with active nematic fluids. These chemicals react and diffuse with each other, and are both advected by the hydrodynamic flow field $\mathbf{u}$ (Eqs.(6.1)–(6.2)). We look at the Schnakenberg model (Eqs.(6.3)–(6.4)) and the Gierer-Meinhardt model (Eqs.(6.3)–(6.4)) in the stripe forming regime. The details of solving these equations numerically are described in Sec. 6.2.3.

B.1.4 Viscoelastic and elastic models

In Chapters 7 and 8, we study materials with an elastic or viscoelastic response to an applied strain.

We consider first in Chapter 7 a purely elastic material, where the stress is proportional to the strain (Eq.(7.4)).

We also use in Chapter 7 the Maxwell model. This model describes a viscoelastic material, which behaves like an elastic solid at short times and a viscous fluid at long times. In this case, the constitutive relation is given by Eq.(7.7), and is solved numerically using a finite-difference method.

Finally, in Chapter 8, we consider a Kelvin-Voigt model. This model describes a viscoelastic material, which behaves opposite to the Maxwell model - it responds

like a viscoelastic fluid at short timescales and an elastic solid at long timescales. In this case, the constitutive relation is different, given by Eq.(8.2).

For all the above models, we have used the coefficients E and η for the elastic modulus and viscosity of the material respectively. They are all solved using finite-difference schemes, and the accuracy of the results are verified using loading and unloading tests, and by numerically verifying the analytical solutions for the stress fields after imposing known flow fields.

B.1.5 Lyotropic model

The lyotropic model uses a single phase field φ to describe different regions of the simulation domain. We have used this model to look at active regions inside passive regions, with $\varphi = 1$ denoting the active region, and $\varphi = 0$ denoting the passive region. The dynamics of the phase field are described by a Cahn-Hilliard equation (Eq.(2.23)). Gradients in φ also give rise to capillary stress in the fluid (Eq.(8.4)), modelling the effects of interfacial surface tension. The dynamics of the phase field are simulated using finite difference.

B.2 Overview of Model Configuration by Chapter

Chapter 3

This chapter studies a two-fluid model, where component 1 is an active nematic and component 2 is a passive isotropic fluid.

Chapter 4

This chapter considers a two-fluid model where both components are active nematics. Sec 4.1 studies director alignment in the absence of phase separation. The remaining sections discuss phase separation when each component fluid has a different activity.

Chapter 5

In this chapter, we consider again a two-fluid model, where component 1 is active and component 2 is passive. This mixture is now placed under confinement.

Chapter 6

This chapter discusses a two chemical fields undergoing reaction and diffusion. The chemicals are also advected by an active flow field, modelled using a single-fluid model. In Sec. 6.4, we further study the case where the activity depends on the local concentration of the chemicals.

Chapter 7

In this chapter, we study a single-fluid lyotropic model with a *fixed* phase field φ labelling different regions. The active region $\varphi = 1$ is inside a channel, confined by the passive region which form the walls of the channel. We study the cases when the passive region $\varphi = 0$ is a purely elastic or a viscoelastic material described by a Maxwell model.

Chapter 8

This chapter considers a single-fluid lyotropic model to study cancer clusters embedded in the ECM. We use a phase field φ to label different domains, which evolves according to the Cahn-Hilliard equation (Eq.(2.23)). The active region $\varphi = 1$ represents cancer clusters. The passive region $\varphi = 0$ represents the viscoelastic ECM, modelled as a Kelvin-Voigt material.

“A true master is an eternal student”

— Master Yi

References

1. S. Bhattacharyya and J. M. Yeomans. Coupling Turing stripes to active flows. *Soft Matter* **17**, 10716–10722 (2021).
2. S. Bhattacharyya and J. M. Yeomans. Phase separation driven by active flows. *Physical Review Letters* **130**, 238201 (2023).
3. F. Mori, S. Bhattacharyya, J. M. Yeomans and S. P. Thampi. Viscoelastic confinement induces periodic flow reversals in active nematics. *Physical Review E* **108**, 064611 (2023).
4. S. Bhattacharyya and J. M. Yeomans. Phase ordering in binary mixtures of active nematic fluids. *Physical Review E* **110**, 024607 (2024).
5. S. Bhattacharyya and J. M. Yeomans. Active nematic phase separation in confined droplets (in preparation).
6. S. Bhattacharyya, P. Gottheil, J. A. Käs, J. M. Yeomans et al. Modelling breast cancer tumours as active nematics in viscoelastic confinement (in preparation).
7. Chen, T., Saw, T. B., Mège, R.-M. & Ladoux, B. Mechanical forces in cell monolayers. *Journal of Cell Science* **131**, jcs218156 (2018).
8. Lee, J., Ishihara, A. & Jacobson, K. How do cells move along surfaces? *Trends in cell biology* **3**, 366–370 (1993).
9. Lazarides, E. & Revel, J. P. The molecular basis of cell movement. *Scientific American* **240**, 100–113 (1979).
10. Angelini, T. E. *et al.* Glass-like dynamics of collective cell migration. *Proceedings of the National Academy of Sciences* **108**, 4714–4719 (2011).
11. Schoetz, E.-M., Lanio, M., Talbot, J. A. & Manning, M. L. Glassy dynamics in three-dimensional embryonic tissues. *Journal of The Royal Society Interface* **10**, 20130726 (2013).
12. Sadati, M., Qazvini, N. T., Krishnan, R., Park, C. Y. & Fredberg, J. J. Collective migration and cell jamming. *Differentiation* **86**, 121–125 (2013).
13. Ranft, J. *et al.* Fluidization of tissues by cell division and apoptosis. *Proceedings of the National Academy of Sciences* **107**, 20863–20868 (2010).
14. Duclut, C., Paijmans, J., Inamdar, M. M., Modes, C. D. & Jülicher, F. Active T1 transitions in cellular networks. *The European Physical Journal E* **45**, 29 (2022).
15. Friedl, P. & Gilmour, D. Collective cell migration in morphogenesis, regeneration and cancer. *Nature Reviews Molecular Cell Biology* **10**, 445–457 (2009).
16. Hogan, B. L. Morphogenesis. *Cell* **96**, 225–233 (1999).

17. Goryachev, A. B. & Mallo, M. Patterning and morphogenesis from cells to organisms: Progress, common principles and new challenges. *Frontiers in cell and developmental biology* **8**, 602483 (2020).
18. Farhadifar, R., Röper, J.-C., Aigouy, B., Eaton, S. & Jülicher, F. The influence of cell mechanics, cell-cell interactions, and proliferation on epithelial packing. *Current biology* **17**, 2095–2104 (2007).
19. Fierling, J. *et al.* Embryo-scale epithelial buckling forms a propagating furrow that initiates gastrulation. *Nature Communications* **13**, 3348 (2022).
20. Webster, G. & Hamilton, S. Budding in hydra: the role of cell multiplication and cell movement in bud initiation. *Development* **27**, 301–316 (1972).
21. De Pascalis, C. & Etienne-Manneville, S. Single and collective cell migration: the mechanics of adhesions. *Molecular biology of the cell* **28**, 1833–1846 (2017).
22. Wang, N. & Ingber, D. E. Control of cytoskeletal mechanics by extracellular matrix, cell shape, and mechanical tension. *Biophysical journal* **66**, 2181–2189 (1994).
23. Friedl, P., Locker, J., Sahai, E. & Segall, J. E. Classifying collective cancer cell invasion. *Nature Cell Biology* **14**, 777–783 (2012).
24. Krakhmal, N. V., Zavyalova, M., Denisov, E., Vtorushin, S. & Perelmuter, V. Cancer invasion: patterns and mechanisms. *Acta Naturae* **7**, 17–28 (2015).
25. Kato, T. *et al.* Interplay of adherens junctions and matrix proteolysis determines the invasive pattern and growth of squamous cell carcinoma. *eLife* **12**, e76520 (2023).
26. Marchetti, M. C. *et al.* Hydrodynamics of soft active matter. *Reviews of Modern Physics* **85**, 1143 (2013).
27. Doostmohammadi, A., Ignés-Mullol, J., Yeomans, J. M. & Sagués, F. Active nematics. *Nature Communications* **9**, 3246 (2018).
28. Jülicher, F., Grill, S. W. & Salbreux, G. Hydrodynamic theory of active matter. *Reports on Progress in Physics* **81**, 076601 (2018).
29. Duclos, G., Erlenkamper, C., Joanny, J.-F. & Silberzan, P. Topological defects in confined populations of spindle-shaped cells. *Nature Physics* **13**, 58–62 (Jan. 2017).
30. Voituriez, R., Joanny, J.-F. & Prost, J. Spontaneous flow transition in active polar gels. *Europhysics Letters* **70**, 404 (2005).
31. Saw, T. B. *et al.* Topological defects in epithelia govern cell death and extrusion. *Nature* **544**, 212–216 (Apr. 2017).
32. Wang, Z., Marchetti, M. C. & Brauns, F. Patterning of morphogenetic anisotropy fields. *Proceedings of the National Academy of Sciences* **120**, e2220167120 (2023).
33. Marenduzzo, D., Orlandini, E., Cates, M. & Yeomans, J. Steady-state hydrodynamic instabilities of active liquid crystals: Hybrid lattice Boltzmann simulations. *Physical Review E* **76**, 031921 (2007).
34. Aditi Simha, R. & Ramaswamy, S. Hydrodynamic Fluctuations and Instabilities in Ordered Suspensions of Self-Propelled Particles. *Physical Review Letters* **89**, 058101 (5 July 2002).

35. Nejad, M. R. & Yeomans, J. M. Spontaneous rotation of active droplets in two and three dimensions. *PRX Life* **1**, 023008 (2023).
36. Turing, A. M. The chemical basis of morphogenesis. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* **237**, 37–72 (1952).
37. Ladoux, B. & Mège, R.-M. Mechanobiology of collective cell behaviours. *Nature Reviews Molecular Cell biology* **18**, 743–757 (2017).
38. Giavazzi, F. *et al.* Flocking transitions in confluent tissues. *Soft Matter* **14**, 3471–3477 (2018).
39. Alert, R., Casademunt, J. & Joanny, J.-F. Active turbulence. *Annual Review of Condensed Matter Physics* **13**, 143–170 (2022).
40. Nonomura, M. Study on multicellular systems using a phase field model. *PloS one* **7**, e33501 (2012).
41. Zhang, G. & Yeomans, J. M. Active forces in confluent cell monolayers. *Physical Review Letters* **130**, 038202 (2023).
42. Honda, H., Tanemura, M. & Nagai, T. A three-dimensional vertex dynamics cell model of space-filling polyhedra simulating cell behavior in a cell aggregate. *Journal of theoretical biology* **226**, 439–453 (2004).
43. Fletcher, A. G., Osterfield, M., Baker, R. E. & Shvartsman, S. Y. Vertex models of epithelial morphogenesis. *Biophysical journal* **106**, 2291–2304 (2014).
44. Bock, M., Tyagi, A. K., Kreft, J.-U. & Alt, W. Generalized Voronoi tessellation as a model of two-dimensional cell tissue dynamics. *Bulletin of mathematical biology* **72**, 1696–1731 (2010).
45. Sánchez-Gutiérrez, D. *et al.* Fundamental physical cellular constraints drive self-organization of tissues. *The EMBO journal* **35**, 77–88 (2016).
46. Haslach Jr, H. W. Nonlinear viscoelastic, thermodynamically consistent, models for biological soft tissue. *Biomechanics and Modeling in Mechanobiology* **3**, 172–189 (2005).
47. Humphrey, J. Stress, strain, and mechanotransduction in cells. *J. Biomech. Eng.* **123**, 638–641 (2001).
48. Godard, B. G. & Heisenberg, C.-P. Cell division and tissue mechanics. *Current opinion in cell biology* **60**, 114–120 (2019).
49. De Gennes, P. & Prost, J. *The Physics of Liquid Crystals* (Clarendon Press, 1993).
50. Onsager, L. The effects of shape on the interaction of colloidal particles. *Annals of the New York Academy of Sciences* **51**, 627–659 (1949).
51. Vink, R. L. Crossover from a Kosterlitz-Thouless phase transition to a discontinuous phase transition in two-dimensional liquid crystals. *Physical Review E* **90**, 062132 (2014).
52. Krueger, T. *et al.* *The Lattice Boltzmann Method: Principles and Practice* (Springer, 2016).
53. Frank, F. C. I. Liquid crystals. On the theory of liquid crystals. *Discussions of the Faraday Society* **25**, 19–28 (1958).

54. Zhang, R., Kumar, N., Ross, J. L., Gardel, M. L. & De Pablo, J. J. Interplay of structure, elasticity, and dynamics in actin-based nematic materials. *Proceedings of the National Academy of Sciences* **115**, E124–E133 (2018).
55. Beris, A. N. & Edwards, B. J. *Thermodynamics of flowing systems: with internal microstructure* **36** (Oxford University Press on Demand, 1994).
56. Bischofs, I. B. & Schwarz, U. S. Cell organization in soft media due to active mechanosensing. *Proceedings of the National Academy of Sciences* **100**, 9274–9279 (2003).
57. Kurpinski, K., Chu, J., Hashi, C. & Li, S. Anisotropic mechanosensing by mesenchymal stem cells. *Proceedings of the National Academy of Sciences* **103**, 16095–16100 (2006).
58. Saw, T. B., Xi, W., Ladoux, B. & Lim, C. T. Biological tissues as active nematic liquid crystals. *Advanced materials* **30**, 1802579 (2018).
59. Killeen, A., Bertrand, T. & Lee, C. F. Polar fluctuations lead to extensile nematic behavior in confluent tissues. *Physical Review Letters* **128**, 078001 (2022).
60. Thampi, S. P., Golestanian, R. & Yeomans, J. M. Vorticity, defects and correlations in active turbulence. *Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences* **372**, 20130366 (2014).
61. Giomi, L., Bowick, M. J., Mishra, P., Sknepnek, R. & Cristina Marchetti, M. Defect dynamics in active nematics. *Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences* **372**, 20130365 (2014).
62. Ramaswamy, S. & Rao, M. Active-filament hydrodynamics: instabilities, boundary conditions and rheology. *New Journal of Physics* **9**, 423–423 (Nov. 2007).
63. Hemingway, E. J., Mishra, P., Marchetti, M. C. & Fielding, S. M. Correlation lengths in hydrodynamic models of active nematics. *Soft Matter* **12**, 7943–7952 (2016).
64. Blow, M. L., Thampi, S. P. & Yeomans, J. M. Biphase, Lyotropic, Active Nematics. *Physical Review Letters* **113**, 248303 (24 Dec. 2014).
65. Lázaro, G. R., Pagonabarraga, I. & Hernández-Machado, A. Phase-field theories for mathematical modeling of biological membranes. *Chemistry and physics of lipids* **185**, 46–60 (2015).
66. Cahn, J. W. & Hilliard, J. E. Free energy of a nonuniform system. I. Interfacial free energy. *The Journal of chemical physics* **28**, 258–267 (1958).
67. Kusumaatmaja, H. & Yeomans, J. M. in *Simulating Complex Systems by Cellular Automata* 241–274 (Springer, 2010).
68. Li, Z., Qiao, Z. & Tang, T. *Numerical solution of differential equations: introduction to finite difference and finite element methods* (Cambridge University Press, 2017).
69. Orszag, S. A. Numerical methods for the simulation of turbulence. *The Physics of Fluids* **12**, II–250 (1969).
70. Marenduzzo, D., Orlandini, E. & Yeomans, J. M. Hydrodynamics and Rheology of Active Liquid Crystals: A Numerical Investigation. *Physical Review Letters* **98**, 118102 (11 Mar. 2007).

71. Dubois, F., Lallemand, P., Obrecht, C. & Mahdi Tekitek, M. Lattice Boltzmann model approximated with finite difference expressions. *Computers & Fluids* **155**. ICMES2015, 3–8 (2017).
72. Hyman, A. A., Weber, C. A. & Jülicher, F. Liquid-Liquid Phase Separation in Biology. *Annual Review of Cell and Developmental Biology* **30**, 39–58 (2014).
73. Banani, S. F., Lee, H. O., Hyman, A. A. & Rosen, M. K. Biomolecular condensates: organizers of cellular biochemistry. *Nature Reviews Molecular Cell Biology* **18**, 285–298 (May 2017).
74. Krens, S. F. & Heisenberg, C.-P. Chapter six - Cell Sorting in Development. *Current Topics in Developmental Biology* **95**, 189–213 (2011).
75. Suzuki, S., Omori, I., Kuraishi, R. & Kaneko, H. Cell sorting and germ layer formation in reconstructed starfish embryos. *Development, Growth & Differentiation* **63**, 343–353 (2021).
76. Sharrock, T. E. & Sanson, B. Cell sorting and morphogenesis in early *Drosophila* embryos. *Seminars in Cell & Developmental Biology* **107**, 147–160 (Nov. 2020).
77. Nadell, C. D., Drescher, K. & Foster, K. R. Spatial structure, cooperation and competition in biofilms. *Nature Reviews Microbiology* **14**, 589–600 (Sept. 2016).
78. Steinberg, M. S. Adhesion-guided multicellular assembly: a commentary upon the postulates, real and imagined, of the differential adhesion hypothesis, with special attention to computer simulations of cell sorting. *Journal of Theoretical Biology* **55**, 431–443 (1975).
79. Foty, R. & Steinberg, M. The differential adhesion hypothesis: a direct evaluation. *Developmental Biology* **278**, 255–263 (Feb. 2005).
80. Brodland, G. W. The Differential Interfacial Tension Hypothesis (DITH): a comprehensive theory for the self-rearrangement of embryonic cells and tissues. *eng. Journal of Biomechanical Engineering* **124**, 188–97 (2 Apr. 2002).
81. Harris, A. K. Is cell sorting caused by differences in the work of intercellular adhesion? A critique of the Steinberg hypothesis. *Journal of Theoretical Biology* **61**, 267–285 (1976).
82. Heine, P., Lippoldt, J., Reddy, G. A., Katira, P. & Käs, J. A. Anomalous cell sorting behavior in mixed monolayers discloses hidden system complexities. *New Journal of Physics* **23**, 043034 (Apr. 2021).
83. Balasubramaniam, L. *et al.* Investigating the nature of active forces in tissues reveals how contractile cells can form extensile monolayers. *Nature Materials* **20**, 1156–1166 (Aug. 2021).
84. Cates, M. E. & Tailleur, J. Motility-induced phase separation. *Annual Review of Condensed Matter Physics* **6**, 219–244 (2015).
85. Gonnella, G., Marenduzzo, D., Suma, A. & Tiribocchi, A. Motility-induced phase separation and coarsening in active matter. *Comptes Rendus Physique* **16**, 316–331 (Apr. 2015).
86. Zhang, J., Alert, R., Yan, J., Wingreen, N. S. & Granick, S. Active phase separation by turning towards regions of higher density. *Nature Physics* **17**, 961 (Aug. 2021).

87. Gokhale, S., Li, J., Solon, A., Gore, J. & Fakhri, N. Dynamic clustering of passive colloids in dense suspensions of motile bacteria. *Physical Review E* **105**, 054605 (5 May 2022).
88. Thutupalli, S., Geyer, D., Singh, R., Adhikari, R. & Stone, H. A. Flow-induced phase separation of active particles is controlled by boundary conditions. *Proceedings of the National Academy of Sciences of the United States of America* **115**, 5403–5408 (May 2018).
89. Furukawa, A., Marenduzzo, D. & Cates, M. E. Activity-induced clustering in model dumbbell swimmers: The role of hydrodynamic interactions. *Physical Review E* **90**, 022303 (2 Aug. 2014).
90. Stewart, H. B. & Wendroff, B. Two-phase flow: models and methods. *Journal of Computational Physics* **56**, 363–409 (1984).
91. Levy, S. *Two-Phase Flow in Complex Systems* (Wiley, 1999).
92. Mogilner, A. & Manhart, A. Intracellular Fluid Mechanics: Coupling Cytoplasmic Flow with Active Cytoskeletal Gel. *Annual Review of Fluid Mechanics* **50**, 347–370 (2018).
93. Weber, C. A., Rycroft, C. H. & Mahadevan, L. Differential Activity-Driven Instabilities in Biphasic Active Matter. *Physical Review Letters* **120**, 248003 (24 June 2018).
94. Cogan, N. G. & Guy, R. D. Multiphase flow models of biogels from crawling cells to bacterial biofilms. *HFSP Journal* **4**, 11–25 (Feb. 2010).
95. Alt, W. & Dembo, M. Cytoplasm dynamics and cell motion: two-phase flow models. *Mathematical Biosciences* **156**, 207–228 (Mar. 1999).
96. Joanny, J. F., Jülicher, F., Kruse, K. & Prost, J. Hydrodynamic theory for multi-component active polar gels. *New Journal of Physics* **9**, 422 (Nov. 2007).
97. Malevanets, A. & M. Yeomans, J. Lattice Boltzmann simulations of complex fluids: viscoelastic effects under oscillatory shear. *Faraday Discuss.* **112**, 237–248 (0 1999).
98. Malevanets, A. & M. Yeomans, J. Modeling viscous drag in binary fluid mixtures. *Computer Physics Communications* **127**, 105–112 (2000).
99. Saffman, P. G. & Taylor, G. I. The penetration of a fluid into a porous medium or Hele-Shaw cell containing a more viscous liquid. *Proceedings of the Royal Society of London. Series A. Mathematical and Physical Sciences* **245**, 312–329 (1958).
100. Pokawanvit, S. *et al.* Active nematic defects in compressible and incompressible flows. *Physical Review E* **106**, 054610 (5 Nov. 2022).
101. Singh, R. & Cates, M. Hydrodynamically interrupted droplet growth in scalar active matter. *Physical Review Letters* **123**, 148005 (2019).
102. Giomi, L. & DeSimone, A. Spontaneous Division and Motility in Active Nematic Droplets. *Physical Review Letters* **112**, 147802 (Apr. 2014).
103. Balasubramaniam, L., Mège, R.-M. & Ladoux, B. Active nematics across scales from cytoskeleton organization to tissue morphogenesis. *Current Opinion in Genetics & Development* **73**, 101897 (2022).

104. Skamrahl, M. *et al.* Cellular segregation in cocultures is driven by differential adhesion and contractility on distinct timescales. eng. *Proceedings of the National Academy of Sciences of the United States of America* **120**, e2213186120 (15 Apr. 2023).
105. Huang, Y. L., Shiau, C., Wu, C., Segall, J. E. & Wu, M. The architecture of co-culture spheroids regulates tumor invasion within a 3D extracellular matrix. *Biophysical reviews and letters* **15**, 131–141 (2020).
106. Shendruk, T. N., Doostmohammadi, A., Thijssen, K. & Yeomans, J. M. Dancing disclinations in confined active nematics. *Soft Matter* **13**, 3853–3862 (2017).
107. Doostmohammadi, A., Shendruk, T. N., Thijssen, K. & Yeomans, J. M. Onset of meso-scale turbulence in active nematics. *Nature Communications* **8**, 15326 (2017).
108. Acheson, D. J. *Elementary fluid dynamics* (Oxford University Press, 1990).
109. Bade, N. D., Kamien, R. D., Assoian, R. K. & Stebe, K. J. Edges impose planar alignment in nematic monolayers by directing cell elongation and enhancing migration. *Soft Matter* **14**, 6867–6874 (33 2018).
110. Doxzen, K. *et al.* Guidance of collective cell migration by substrate geometry. *Integrative Biology* **5**, 1026–1035 (June 2013).
111. Alert, R. Fingering instability of active nematic droplets. *Journal of Physics A: Mathematical and Theoretical* **55**, 234009 (May 2022).
112. Opatthalage, A. *et al.* Self-organized dynamics and the transition to turbulence of confined active nematics. *Proceedings of the National Academy of Sciences* **116**, 4788–4797 (2019).
113. Graham, J. N., Zhang, G. & Yeomans, J. M. Cell sorting by active forces in a phase-field model of cell monolayers. *Soft Matter* (2024).
114. Krajnc, M. Solid–fluid transition and cell sorting in epithelia with junctional tension fluctuations. *Soft Matter* **16**, 3209–3215 (13 2020).
115. Hallatschek, O. *et al.* Proliferating active matter. *Nature Reviews Physics* **5**, 407–419 (July 2023).
116. Dell’Arciprete, D. *et al.* A growing bacterial colony in two dimensions as an active nematic. *Nature Communications* **9**, 4190 (Oct. 2018).
117. Agudo-Canalejo, J. & Golestanian, R. Active Phase Separation in Mixtures of Chemically Interacting Particles. *Physical Review Letters* **123**, 018101 (1 July 2019).
118. Gross, P., Kumar, K. V. & Grill, S. W. How Active Mechanics and Regulatory Biochemistry Combine to Form Patterns in Development. *Annual Review of Biophysics* **46**. PMID: 28532214, 337–356 (2017).
119. Wyczalkowski, M. A., Chen, Z., Filas, B. A., Varner, V. D. & Taber, L. A. Computational models for mechanics of morphogenesis. eng. *Birth defects research. Part C, Embryo today : reviews* **96**. PMC3374861[pmcid], 132–152 (June 2012).
120. Gierer, A. & Meinhardt, H. A theory of biological pattern formation. *Kybernetik* **12**, 30–39 (Dec. 1972).
121. Kondo, S. & Miura, T. Reaction-Diffusion Model as a Framework for Understanding Biological Pattern Formation. *Science* **329**, 1616–1620 (2010).

122. Howard, J., Grill, S. W. & Bois, J. S. Turing's next steps: the mechanochemical basis of morphogenesis. *Nature Reviews Molecular Cell Biology* **12**, 392–398 (June 2011).
123. Landge, A. N., Jordan, B. M., Diego, X. & Müller, P. Pattern formation mechanisms of self-organizing reaction-diffusion systems. *Developmental Biology* **460**. Systems Biology of Pattern Formation, 2–11 (2020).
124. Yin, S., Li, B. & Feng, X.-Q. Bio-chemo-mechanical theory of active shells. *Journal of the Mechanics and Physics of Solids* **152**, 104419 (2021).
125. Recho, P., Hallou, A. & Hannezo, E. Theory of mechanochemical patterning in biphasic biological tissues. *Proceedings of the National Academy of Sciences* **116**, 5344–5349 (2019).
126. Maini, P. K., Woolley, T. E., Baker, R. E., Gaffney, E. A. & Lee, S. S. Turing's model for biological pattern formation and the robustness problem. *Interface Focus* **2**, 487–496 (2012).
127. Siero, E. *et al.* Striped pattern selection by advective reaction-diffusion systems: Resilience of banded vegetation on slopes. *Chaos: An Interdisciplinary Journal of Nonlinear Science* **25**, 036411 (2015).
128. Bois, J. S., Jülicher, F. & Grill, S. W. Pattern Formation in Active Fluids. *Physical Review Letters* **106**, 028103 (2 Jan. 2011).
129. Kumar, K. V., Bois, J. S., Jülicher, F. & Grill, S. W. Pulsatory patterns in active fluids. *Physical Review Letters* **112**, 208101 (2014).
130. Mietke, A., Jülicher, F. & Sbalzarini, I. F. Self-organized shape dynamics of active surfaces. *Proceedings of the National Academy of Sciences* **116**, 29–34 (2019).
131. Schnakenberg, J. Simple chemical reaction systems with limit cycle behaviour. *Journal of Theoretical Biology* **81**, 389–400 (1979).
132. Liu, P., Shi, J., Wang, Y. & Feng, X. Bifurcation analysis of reaction–diffusion Schnakenberg model. *Journal of Mathematical Chemistry* **51**, 2001–2019 (Sept. 2013).
133. Dufiet, V. & Boissonade, J. Conventional and unconventional Turing patterns. *The Journal of Chemical Physics* **96**, 664–673 (1992).
134. Murray, J. Parameter space for turing instability in reaction diffusion mechanisms: A comparison of models. *Journal of Theoretical Biology* **98**, 143–163 (1982).
135. Gray, P. & Scott, S. K. Sustained oscillations and other exotic patterns of behavior in isothermal reactions. *The Journal of Physical Chemistry* **89**, 22–32. eprint: <https://doi.org/10.1021/j100247a009> (1985).
136. Grindrod, P. The theory and applications of reaction-diffusion equations: patterns and waves. (*No Title*) (1996).
137. Thampi, S. P. & Yeomans, J. M. Active turbulence in active nematics. *The European Physical Journal Special Topics* **225**, 651–662 (2016).
138. in. *Combustion: Physical and Chemical Fundamentals, Modeling and Simulation, Experiments, Pollutant Formation* 187–212 (Springer Berlin Heidelberg, Berlin, Heidelberg, 2006).

139. Rickert, W., Vilchevskaya, E. N. & Müller, W. H. A note on couette flow of nematic crystals according to the Ericksen–Leslie theory. *ZAMM - Journal of Applied Mathematics and Mechanics / Zeitschrift für Angewandte Mathematik und Mechanik* **100**, e201900035 (2020).
140. Leslie, F. M. in *Theory and Applications of Liquid Crystals* (eds Ericksen, J. L. & Kinderlehrer, D.) 235–254 (Springer New York, New York, NY, 1987).
141. Van Horn, B. L. & Winter, H. H. Dynamics of shear aligning of nematic liquid crystal monodomains. *Rheologica Acta* **39**, 294–300 (Aug. 2000).
142. Bonelli, F. *et al.* Lamellar ordering, droplet formation and phase inversion in exotic active emulsions. *Scientific Reports* **9**, 2801 (Feb. 2019).
143. Negro, G., Carenza, L. N., Lamura, A., Tiribocchi, A. & Gonnella, G. Rheology of active polar emulsions: from linear to unidirectional and inviscid flow, and intermittent viscosity. *Soft Matter* **15**, 8251–8265 (41 2019).
144. Hannezo, E., Dong, B., Recho, P., Joanny, J.-F. & Hayashi, S. Cortical instability drives periodic supracellular actin pattern formation in epithelial tubes. *Proceedings of the National Academy of Sciences* **112**, 8620–8625 (2015).
145. Lecuit, T. & Lenne, P.-F. Cell surface mechanics and the control of cell shape, tissue patterns and morphogenesis. *Nature reviews Molecular cell biology* **8**, 633–644 (2007).
146. Salbreux, G., Charra, G. & Paluch, E. Actin cortex mechanics and cellular morphogenesis. *Trends in cell biology* **22**, 536–545 (2012).
147. Sunyer, R. & Treppe, X. Durotaxis. *Current Biology* **30**, R383–R387 (2020).
148. Conrad, J. C. & Poling-Skutvik, R. Confined flow: consequences and implications for bacteria and biofilms. *Annual Review of Chemical and Biomolecular Engineering* **9**, 175–200 (2018).
149. Patteson, A., Gopinath, A., Goulian, M. & Arratia, P. Running and tumbling with *E. coli* in polymeric solutions. *Scientific Reports* **5**, 15761 (2015).
150. Zöttl, A. & Yeomans, J. M. Enhanced bacterial swimming speeds in macromolecular polymer solutions. *Nature Physics* **15**, 554–558 (2019).
151. Chaudhuri, O., Cooper-White, J., Janmey, P. A., Mooney, D. J. & Shenoy, V. B. Effects of extracellular matrix viscoelasticity on cellular behaviour. *Nature* **584**, 535–546 (2020).
152. Clark, A. G. *et al.* Self-generated gradients steer collective migration on viscoelastic collagen networks. *Nature Materials* **21**, 1200–1210 (2022).
153. Elosegui-Artola, A. *et al.* Matrix viscoelasticity controls spatiotemporal tissue organization. *Nature Materials* **22**, 117–127 (2023).
154. Style, R. W. *et al.* Traction force microscopy in physics and biology. *Soft Matter* **10**, 4047–4055 (2014).
155. Colin-York, H. & Fritzsche, M. The future of traction force microscopy. *Current Opinion in Biomedical Engineering* **5**, 1–5 (2018).
156. Deforet, M., Hakim, V., Yevick, H., Duclos, G. & Silberzan, P. Emergence of collective modes and tri-dimensional structures from epithelial confinement. *Nature Communications* **5**, 3747 (2014).

157. Wioland, H., Lushi, E. & Goldstein, R. E. Directed collective motion of bacteria under channel confinement. *New Journal of Physics* **18**, 075002 (2016).
158. Chandrakar, P. *et al.* Confinement controls the bend instability of three-dimensional active liquid crystals. *Physical Review Letters* **125**, 257801 (2020).
159. Singh, A., Vagne, Q., Jülicher, F. & Sbalzarini, I. F. Three Dimensional Spontaneous Flow Transition in Active Polar Fluids. *arXiv preprint arXiv:2302.04259* (2023).
160. Giomi, L., Mahadevan, L., Chakraborty, B. & Hagan, M. Banding, excitability and chaos in active nematic suspensions. *Nonlinearity* **25**, 2245 (2012).
161. Hardoüin, J. *et al.* Reconfigurable flows and defect landscape of confined active nematics. *Communications Physics* **2**, 121 (2019).
162. Thampi, S. P. Channel Confined Active Nematics. *Current Opinion in Colloid & Interface Science*, 101613 (2022).
163. Chandragiri, S., Doostmohammadi, A., Yeomans, J. M. & Thampi, S. P. Active transport in a channel: stabilisation by flow or thermodynamics. *Soft Matter* **15**, 1597–1604 (2019).
164. Samui, A., Yeomans, J. M. & Thampi, S. P. Flow transitions and length scales of a channel-confined active nematic. *Soft Matter* **17**, 10640–10648 (2021).
165. Joshi, C. *et al.* From disks to channels: Dynamics of active nematics confined to an annulus. *arXiv preprint arXiv:2304.04895* (2023).
166. Araújo, N. A. *et al.* Steering self-organisation through confinement. *Soft Matter* **19**, 1695–1704 (2023).
167. Jülicher, F., Kruse, K., Prost, J. & Joanny, J.-F. Active behavior of the cytoskeleton. *Physics Reports* **449**, 3–28 (2007).
168. Marcq, P. Spatio-temporal dynamics of an active, polar, viscoelastic ring. *The European Physical Journal E* **37**, 1–8 (2014).
169. Li, G. & Ardekani, A. M. Collective motion of microorganisms in a viscoelastic fluid. *Physical Review Letters* **117**, 118001 (2016).
170. Hemingway, E. J., Cates, M. E. & Fielding, S. M. Viscoelastic and elastomeric active matter: linear instability and nonlinear dynamics. *Physical Review E* **93**, 032702 (2016).
171. Liu, S., Shankar, S., Marchetti, M. C. & Wu, Y. Viscoelastic control of spatiotemporal order in bacterial active matter. *Nature* **590**, 80–84 (2021).
172. De Kinkelder, E. M., Fischer-Friedrich, E. & Aland, S. Pulsatory patterns in active viscoelastic fluids with distinct relaxation time scales. *New Journal of Physics* (2023).
173. Emmanuel, L., Yeomans, J. M. & Doostmohammadi, A. Active matter in a viscoelastic environment. *Physical Review Fluids* **5**, 023102 (2020).
174. Plan, E. L. V. M., Yeomans, J. M. & Doostmohammadi, A. Activity pulses induce spontaneous flow reversals in viscoelastic environments. *Journal of the Royal Society Interface* **18**, 20210100 (2021).

175. Emmanuel, L., Le Thi, H., Yeomans, J. M. & Doostmohammadi, A. Self-sustained oscillations of active viscoelastic matter. *Journal of Physics A: Mathematical and Theoretical* **55**, 275601 (2022).
176. Hamby, A. E., Vig, D. K., Safonova, S. & Wolgemuth, C. W. Swimming bacteria power microspin cycles. *Science Advances* **4**, eaau0125 (2018).
177. Landau, L. D., Lifshitz, E. M., Kosevich, A. M. & Pitaevskii, L. P. *Theory of elasticity: volume 7* (Elsevier, 1986).
178. Joseph, D. D. *Fluid dynamics of viscoelastic liquids* (Springer Science & Business Media, 2013).
179. Slaughter, W. S. *The linearized theory of elasticity* (Springer Science & Business Media, 2012).
180. Maxwell, J. C. IV. On the dynamical theory of gases. *Philosophical transactions of the Royal Society of London*, 49–88 (1867).
181. Thomson, W. IV. On the elasticity and viscosity of metals. *Proceedings of the Royal Society of London*, 289–297 (1865).
182. Voigt, W. Ueber innere Reibung fester Körper, insbesondere der Metalle. *Annalen der Physik* **283**, 671–693 (1892).
183. Oldroyd, J. G. On the formulation of rheological equations of state. *Proceedings of the Royal Society of London. Series A. Mathematical and Physical Sciences* **200**, 523–541 (1950).
184. Bonfanti, A., Kaplan, J. L., Charras, G. & Kabla, A. Fractional viscoelastic models for power-law materials. *Soft Matter* **16**, 6002–6020 (2020).
185. Wagner, A. J. The origin of spurious velocities in Lattice Boltzmann. *International Journal of Modern Physics B* **17**, 193–196 (2003).
186. Guo, Z., Zheng, C. & Shi, B. Discrete lattice effects on the forcing term in the lattice Boltzmann method. *Physical Review E* **65**, 046308 (Apr. 2002).
187. Wood, W., Muss, H. & Solin, L. *Olopade Ol. Malignant tumors of the breast. In: DeVita VT, Hellman S, Rosenberg S A. Cancer. 7, h ed 2005.*
188. Łukasiewicz, S. *et al.* Breast cancer—epidemiology, risk factors, classification, prognostic markers, and current treatment strategies—an updated review. *Cancers* **13**, 4287 (2021).
189. Scully, O. J., Bay, B.-H., Yip, G. & Yu, Y. Breast cancer metastasis. *Cancer genomics & proteomics* **9**, 311–320 (2012).
190. Janowczyk, A. & Madabhushi, A. Deep learning for digital pathology image analysis: A comprehensive tutorial with selected use cases. *Journal of pathology informatics* **7**, 29 (2016).
191. Villa, C., Chaplain, M. A., Gerisch, A. & Lorenzi, T. Mechanical models of pattern and form in biological tissues: The role of stress–strain constitutive equations. *Bulletin of Mathematical Biology* **83**, 80 (2021).
192. Rajagopal, K. R. A note on a reappraisal and generalization of the Kelvin–Voigt model. *Mechanics research communications* **36**, 232–235 (2009).

193. Mège, R. M. Molecular basis for fluidization of cancer cells. *Nature Materials* **18**, 1147–1148 (2019).
194. Alcaraz, J. *et al.* Collective epithelial cell invasion overcomes mechanical barriers of collagenous extracellular matrix by a narrow tube-like geometry and MMP14-dependent local softening. *Integrative biology* **3**, 1153–1166 (2011).
195. Acerbi, I. *et al.* Human breast cancer invasion and aggression correlates with ECM stiffening and immune cell infiltration. *Integrative Biology* **7**, 1120–1134 (2015).
196. Gottheil, P. *et al.* State of cell unjamming correlates with distant metastasis in cancer patients. *Physical Review X* **13**, 031003 (2023).
197. Grosser, S. *et al.* Cell and nucleus shape as an indicator of tissue fluidity in carcinoma. *Physical Review X* **11**, 011033 (2021).
198. Liew, H. Y. *et al.* Cellular Traction Force Holds the Potential as a Drug Testing Readout for In Vitro Cancer Metastasis. *Cellular and Molecular Bioengineering* **17**, 203–217 (2024).
199. Kraning-Rush, C. M., Califano, J. P. & Reinhart-King, C. A. Cellular traction stresses increase with increasing metastatic potential. *PloS one* **7**, e32572 (2012).
200. Weibel, E. R., Kistler, G. S. & Scherle, W. F. Practical stereological methods for morphometric cytology. *The Journal of cell biology* **30**, 23–38 (1966).
201. Wicksell, S. D. The Corpuscle Problem: A Mathematical Study of a Biometric Problem. *Biometrika* **17**, 84–99 (1925).
202. Wicksell, S. D. The Corpuscle Problem: Second Memoir: Case of Ellipsoidal Corpuscles. *Biometrika* **18**, 151–172 (1926).
203. Sahagian, D. L. & Proussevitch, A. A. 3D particle size distributions from 2D observations: stereology for natural applications. *Journal of Volcanology and Geothermal Research* **84**, 173–196 (1998).
204. Macenko, M. *et al.* A method for normalizing histology slides for quantitative analysis in 2009 IEEE international symposium on biomedical imaging: from nano to macro (2009), 1107–1110.
205. Marusyk, A. & Polyak, K. Tumor heterogeneity: causes and consequences. *Biochimica et Biophysica Acta (BBA)-Reviews on Cancer* **1805**, 105–117 (2010).
206. Frost, I. *et al.* Cooperation, competition and antibiotic resistance in bacterial colonies. *The ISME journal* **12**, 1582–1593 (2018).
207. Yamagishi, J. F., Saito, N. & Kaneko, K. Symbiotic cell differentiation and cooperative growth in multicellular aggregates. *PLoS computational biology* **12**, e1005042 (2016).
208. Ruske, L. J. & Yeomans, J. M. Activity gradients in two- and three-dimensional active nematics. *Soft Matter* **18**, 5654–5661 (30 2022).
209. Mongera, A. *et al.* A fluid-to-solid jamming transition underlies vertebrate body axis elongation. *Nature* **561**, 401–405 (2018).
210. Patil, P. U., D'Ambrosio, J., Inge, L. J., Mason, R. W. & Rajasekaran, A. K. Carcinoma cells induce lumen filling and EMT in epithelial cells through soluble E-cadherin-mediated activation of EGFR. *Journal of cell science* **128**, 4366–4379 (2015).

- 211. Thampi, S. P., Golestanian, R. & Yeomans, J. M. Vorticity, defects and correlations in active turbulence. *Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences* **372**, 20130366 (2014).
- 212. Chapman, S. & Cowling, T. G. *The mathematical theory of non-uniform gases: an account of the kinetic theory of viscosity, thermal conduction and diffusion in gases* (Cambridge University Press, 1990).