Collective cell migration of epithelial cells driven by chiral torque generation

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(Dated: September 9, 2019)

Various multicellular tissues show chiral morphology. Experimental studies have shown this can originate from cell chirality. However, no theory has been proposed to connect the cellular chiral torque and multicellular chiral morphogenesis. We propose a model of confluent tissue dynamics with cellular chiral torque. We found that cells migrate unidirectionally under a gradient of cellular chiral torque. While the migration speed varies depending on the tissue’s mechanical parameters, it is scaled solely by a structural order parameter for liquid-to-solid transition in confluent tissues.

The establishment of left-right (LR) asymmetry in tissues and organs is an intriguing event in development, which involves the coordinated activity of cells and molecules [1–4]. Since proteins as the basic components of biological systems are chiral molecules, the breaking of LR symmetry can be a collective property orchestrated by these chiral molecular components [2, 5].

For the LR asymmetry in tissue morphogenesis, it has been shown that in Drosophila, the embryonic hindgut twists unidirectionally [7], and the male genitalia undergoes a clockwise rotation when looked from the apical side [8, 9]. These are epithelial tissues, which consist of epithelial cells that adhere to each other through cell-cell junctions [10]. During the hindgut twisting, the hindgut epithelial cells exhibit chiral properties in their shape and other properties [2]. In the genitalia rotation, the surrounding epithelial cells move collectively in the clockwise direction driven by LR asymmetric positional rearrangements of cells, which rotates the genital disk [8, 11]. Even more generally, such chiral cell rearrangements can induce unidirectional cell migration within an epithelial tissue [12]. The above examples suggest that a collective behavior of cellular chirality gives rise to the tissue LR asymmetry.

At the single cell level, several types of cells have been reported to exhibit chiral dynamics. Examples include the extension of neurites in cultured nerve cell [13], the chiral movement of C. elegans cell cortex [14], nuclear rotation in melanophores of zebrafish [15] and actin cytoskeleton swirling in human foreskin fibroblasts (HFFs) cultured on a micro pattern [16]. Remarkably, in zebrafish melanophores and the HFFs on the micro pattern, the single cells can cell-autonomously generate chiral torques in such a way that the apical side (top side) of cells exhibits rotations with respect to the basal side (bottom side) which adheres to substrates. Although a variety of specific mechanisms generate chiral torque at the cellular level, many of these chiral properties are governed by active chiral processes of actomyosin cytoskeleton [17–19].

Since actomyosin is a ubiquitous component of eukaryotic cells, it is natural to consider that chiral torque generation is not a feature restricted to the specific cells mentioned above. In particular, we consider a situation where chiral torque is generated by individual single epithelial cells in a tissue, motivated by the LR symmetry breaking of the epithelial tissues [12, 13], and by the fact that single cells can generate chiral torque [13–16]. Based on the cell vertex model (CVM) [20], we propose a theoretical model of dynamics of an epithelial tissue with a chiral torque generated by individual single cells. Then we investigate the tissue scale dynamical emergent properties.

To model the dynamics of an epithelial tissue with chiral torques generated by individual single cells, we use a two dimensional (2D) CVM. In the CVM, cells are described by polygons with vertices and edges. The position of the ith vertex is represented by $\mathbf{r}_i$. The force acting on each vertex is given by the derivative $-\partial E(\{\mathbf{r}_i\})/\partial \mathbf{r}_i$ of a potential function $E(\{\mathbf{r}_i\})$, given by

$$E(\{\mathbf{r}_i\}) = \frac{K}{2} \sum_{\alpha=1}^{N} (A_\alpha - A_0)^2 + \frac{K_p}{2} \sum_{\alpha=1}^{N} (P_\alpha - P_0)^2 + \sum_{\langle i,j \rangle} \Delta A_{ij}(t) \ell_{ij}. \quad (1)$$

Here, the first term on the right hand side describes the area elasticity with the area $A_\alpha$ of cell $\alpha$, the elastic modulus $K$, and the preferred area $A_0$. The second term defines the perimeter elasticity with the perimeter length $P_\alpha$ of cell $\alpha$, the elastic modulus $K_p$, and the preferred perimeter length $P_0$. We assume for simplicity that these cellular mechanical properties $K, K_p, A_0, P_0$ are spatially homogeneous. $N$ is the total number of cells. The third term is introduced to explicitly describe the fluctuation in the line tension $\Delta A_{ij}(t)$. Here, $\ell_{ij}$ is the length of a cell edge between ith and jth vertices. We introduce a fluctuating tension $\Delta A_{ij}(t)$ as a colored Gaussian noise with $\langle \Delta A_{ij}(t) \rangle = 0$ and $\langle \Delta A_{ij}(t_1) \Delta A_{kl}(t_2) \rangle = \delta_{ik}\delta_{jl}\delta^{2}e^{-|t_1-t_2|/\tau}$. Such time-correlated fluctuation of line tension is reported in [21]. We set $\tau = 1$ in this letter for simplicity. Hereafter, we choose the units of length and forces such that the elastic
where \( \nu \) by the cell \( \alpha \) and square root of area \([22, 23]\). If a single cell tends to target shape index, which is a ratio between perimeter \( K \) and the preferred cell area \( A_0 \). We set \( K_0 = 3.54 \) and \( L = 10 \) to avoid large cellular shape deformation unless otherwise noted.

In the epithelial tissue, we also consider that a torque driven by the torque generated by actomyosin network.

\[
\bar{\mathbf{T}}_i = \sum_{\alpha \text{cell}} \nu_\alpha (\bar{r}_i - \bar{r}_g) \times \bar{n},
\]

where \( \nu_\alpha \) is the coefficient of the torque force generated by the cell \( \alpha \), \( \bar{r}_g \) is the area centroid of cell \( \alpha \), and \( \bar{n} \) is a unit normal vector from the basal to apical sides.

The time evolution equation for \( \bar{r}_i \) is obtained by considering the force balance between the frictional force \( \eta d\bar{r}_i/dt \) with friction constant \( \eta \), the potential force \( -\partial E(\{\bar{r}_i\})/\partial \bar{r}_i \) derived from Eq. (1), and the torque force \( \bar{T}_i \) given by Eq. (2) as follows:

\[
\eta \frac{d\bar{r}_i}{dt} = -\frac{\partial E(\{\bar{r}_i\})}{\partial \bar{r}_i} + \bar{T}_i.
\]

When the length of a cell edge falls below a threshold \( l_{th} = 0.03 \) during a time-evolution according to Eq. (3), a T1 transition is performed by flipping the edge by 90°.

We consider a simple configuration of a rectangular epithelial sheet with size \( L_x \) and \( L_y \) in the \( x \)- and \( y \)-direction, respectively, as shown in Fig. 1(b). For the \( x \)-direction, we apply the periodic boundary condition at \( x = 0 \) and \( x = L_x \). The rectangular area size is equal to the number of cells so that the average cell area is set to be \( A_0 = 1 \). This configuration corresponds to the tube geometry, a biologically ubiquitous structure of organs, which exhibits chiral morphogenesis as twisting of a heart tube and hindgut. We simply consider that the cells are attached to the boundary. We hence fix the \( y \)-coordinates of vertices on the bottom and top boundaries to \( y = 0 \) and \( y = L_y \), respectively. \( \eta \) is set to 1 unless otherwise noted. We calculate the time-evolution Eq. (3) with the time step \( \Delta t = 0.01 \). We prepare initial cellular configurations packed with regular hexagonal cells to calculate the dynamics with \( \sigma = 0 \) and \( \nu_\alpha = 0 \) to relax the system, and then set \( \sigma \) and \( \nu_\alpha \) to the target values. The numbers of cells in the initial hexagonal configuration in the \( x \)- and \( y \)-directions are respectively denoted as \( N_x \) and \( N_y \), and we set \( N_x = 10 \) for all the simulation.

We first investigated the role of chiral torque generation on the dynamics of cells when the strength \( \nu_\alpha \) of chiral torque generation is spatially homogeneous. As shown in Fig. 2(a), we found that the cells migrate bidirectionally when the fluctuation in line tension is present (\( \nu_\alpha = 0.2, \sigma = 0.3, N_y = 6 \)). With negative \( \nu_\alpha \), the cellular behavior was reversed, confirming that the chiral torque generation is the driving force of the cellular migration. For sufficiently large system (\( N_y = 40 \)), the flow profile rapidly decays near the boundary, and the bulk velocity vanishes (Fig. 2(b)). This indicates that the torque force and the potential force are balanced in
the bulk, while not balanced near the boundary. On the
top and bottom boundaries, the torque forces on the ver-
tices are exerted in the rightward and leftward direction,
respectively, as depicted in Fig. 2(c), leading to the bidi-
rectional cellular flow at the boundaries.

In the bulk, to understand how the torque force and
the potential force are balanced, we consider a mean-
field model where the torque force is exerted on a vertex
O surrounded by three regular hexagonal cells as shown
in Fig. 1(a), without the line tension fluctuation. When
να is homogeneous, we readily find that the torque force
exerted on the vertex O vanishes due to the 3-fold sym-
metry. Therefore, even though the torque force is present
in the bulk due to cellular deformation, it should be so
small that the deformation of cells can readily balance it.

We note that without the line tension fluctuation (σ = 0),
all the cells are just deformed in the asymmetric fashion
without continuous cellular flow (See Supplemental
Material 24)). Hence, the cell rearrangements induced
by the stochastic fluctuation of the line tension promote
the continuous cellular flow.

We next consider a condition in which collective cell
migration appears in a defined direction, induced by chiral
torque generation. According to a symmetry argument
12, the symmetry along the y-axis has to be bro-
en to achieve a unidirectional cellular movement along
the x-axis. In this letter, we consider a situation where
chiral torque generation depends on the position of the
cells along the y-axis. We simply assume a linear form
given by να = −λ(yα − Lα), where yα is the y-coordinate
of the center of cell α. Here, we impose a boundary con-
dition in which the friction coefficients on the top and
bottom boundaries are considerably higher (η = 10000)
to avoid the effect of the boundary torque force as we
discussed above (Fig. 2(c)). With the torque gradi-
ent, we found that cells migrate unidirectionally in the
direction perpendicular to that of the torque gradient
(Fig. 3a)). A time-averaged flow profile is shown in
Fig. 3b) (λ = 0.01, σ = 0.3, Ny = 40). With negative λ, the direction of the cellular migration is reversed. As λ increases, the steady-state cellular velocity V_{ave} in the x-axis increases almost linearly (Fig. 3c)). These re-
results confirm that the chiral torque gradient is the driving
force for this unidirectional collective cell migration.
We also found that without the line tension fluctuation, the
cells only deform without continuous flow (See Supple-
mental Material 24)). Hence, the cell rearrangements
induced by the stochastic fluctuation in the line tension
are necessary for the continuous cellular flow.

In contrast to the case in which the strength of
torque strength is homogeneous, the flow profile shown in
Fig. 3b) indicates that the cells can migrate in the
bulk region under the torque gradient. To see the mecha-

nism of the unidirectional cellular migration, we con-
der the mean field model of three cells (Fig. 4a)). We
set the position of the target vertex O as the origin of

![Figure 3](image-url)

**FIG. 3.** (a) Time evolution of a cellular configuration in a
numerical simulation (λ = 0.1, σ = 0.3, N_y = 6). The time
interval between each configuration is 40 time units. Sup-
plemental Material S2 [24] provides the movie. (b) Time-
averaged flow profile for N_y = 40 averaged over 5 samples
(λ = 0.01, σ = 0.3). (c) Average velocity V_{ave} of cells is plotted
against torque gradient λ for different intensities σ
(N_y = 6). The error bars represent the standard deviation.
found that an increase of either $P_0$ or $\sigma$ increases (inset of Fig. 4). To see the influence of $P_0$ and $\sigma$ in a unified way, we pay attention to the cell shape $q_\alpha$ obtained from the numerical simulations. In Fig. 5 we plotted the migration velocity $V_{bulk}$ in the bulk layers against the cell shape $q_\alpha$ averaged in the boundary layers, and found that the velocity curves in the inset of Fig. 5 are surprisingly collapsed in a single line. Hence, $\langle \alpha, \langle q_\alpha \rangle_{BL} \rangle$ is an excellent indicator of the migration velocity in our model. Here, we define the boundary and bulk layers from the flow profile, and exclude the cells on the bottom and top boundaries for calculation of $\langle \alpha, \langle q_\alpha \rangle_{BL} \rangle$, since the cells are strongly deformed by the flat boundaries (See Supplemental Material 24).

We discuss how the bulk velocity $V_{bulk}$ is uniquely determined by the cell shape $\langle q_\alpha \rangle_{BL}$, in Fig. 5. The cellular velocity should be determined by the balance between the bulk torque force and how easily cells rearrange over the energy barrier of T1 transition in the boundary layers. In the present model, since the torque force depends on the cell shape, the bulk torque force should be determined by the cell shape $\langle q_\alpha \rangle_{BU}$ averaged in the bulk. The cell shape $q_\alpha$ has been also suggested as an indicator of how easily cells rearrange over the energy barrier of T1 transition 23,24. Hence, the energy barrier of T1 transition in the boundary layers depends on $\langle \alpha, \langle q_\alpha \rangle_{BL} \rangle$. Consequently, considering that $\langle \alpha, \langle q_\alpha \rangle_{BL} \rangle \approx \langle \alpha, \langle q_\alpha \rangle_{BU} \rangle$ is satisfied (See Supplemental Material 24), and hence both bulk torque force and energy barrier of T1 transition depend on $\langle \alpha, \langle q_\alpha \rangle_{BL} \rangle$, $\langle \alpha, \langle q_\alpha \rangle_{BL} \rangle$ uniquely determines the migration velocity.

In this letter, we have reported that a dynamical chiral property at the single-cell level, which is cellular chiral torque generation, can induce the collective cellular migration. Our model predicts that, when the strength of torque is spatially homogeneous, the torque forces in the bulk almost balanced, and those generated by the cells at the boundaries drive the bidirectional cellular migration. Another prediction is that, under the gradient of chiral torque strength, the cells can migrate unidirectionally and perpendicularly to the gradient driven by the torque force generated in the bulk. Although the mechanism of the torque generation at the single-cell level is not fully revealed experimentally, previous studies reported that a combination of cytoskeleton and motor proteins generates the cellular torque. Activity of such cytoskeleton and motor proteins is regulated by various biochemical pathways such as Rho signaling pathway 27. Hence, the spatial gradient of regulatory molecules should produce a gradient of the strength of the cellular torque. In in vivo systems, an epithelial tissue is attached to other different tissues, and hence such attached tissues probably emit biochemical signals to generate the concentration gradient of molecules regulating the strength of the cellular
torque. Since both the cellular chiral torque generation and the gradient of the cellular torque are considered to be ubiquitous in biological systems, we expect that the mechanism of the LR symmetry breaking in tissue dynamics we have proposed plays an essential role in the chiral collective cellular movements, such as a twist of epithelial tube [7] and a unidirectional epithelial cellular flow [8], during development.

This work is supported by Grant-in-Aid for JSPS Fellows (Grant No. 18J01239 to TY), KAKENHI Grant No. 17H07366 (to TY), JP16K17777 (to TH), JP19K03764 (to TH) and JP19H00996 (to TS), and JST CREST grant number JPMJCR1852, Japan (TS).

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SUPPLEMENTAL MATERIAL
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I. DEFINITION OF THE AREA CENTROID OF A CELL

We define the cell center of cell \( \alpha \) using the area centroid as \( \vec{r}_g^{\alpha} = \frac{1}{6A_\alpha} \sum_{\mu=1}^{N_\alpha} (\vec{r}_\mu^{\alpha} + \vec{r}_{N_\alpha+1}^{\alpha}) (\vec{r}_\mu^{\alpha} \times \vec{r}_{N_\alpha+1}^{\alpha}) \cdot \vec{n} \). Here, \( \mu \) is the index of the \( N_\alpha \) vertices around the cell \( \alpha \), indexed in the counterclockwise direction. The position vector of each vertex is represented by \( \vec{r}_\mu^{\alpha} \) and \( \vec{r}_{N_\alpha+1}^{\alpha} = \vec{r}_1^{\alpha} \).

II. PREPARATION OF THE INITIAL CELLULAR CONFIGURATION

We prepared the initial condition as shown in Fig. S1(a). In the initial condition, the cells in the bulk are set to regular hexagonal shape, while the cells at the boundaries, which are labeled with red dots in Fig. S1(a), are set to half of the regular hexagonal cells. The average cell area \( \bar{A} \) is set to 1. Then, we calculate the dynamics with \( \sigma = 0 \) and \( \nu_\alpha = 0 \) to relax the system for 50 time units, and then we set \( \sigma \) and \( \nu_\alpha \) to the target values. Fig. S1(b) is the configuration after the relaxation (\( P_0 = 3.54 \)).

![Fig. S1. (a) The initial condition with regular hexagonal cells. The half regular hexagonal cells on the boundaries are shown with red dots. (b) A configuration after relaxation with \( \sigma = 0 \) and \( \nu_\alpha = 0 \) (\( P_0 = 3.54 \)).](image)

III. CONFIGURATION OBTAINED WHEN \( \sigma = 0 \).

![Fig. S2. The tissue deformation observed without continuous cellular flow in the zero-noise limit \( \sigma = 0 \) (a) under a homogeneous cellular chiral torque (\( \nu_\alpha = 0.2 \)) and (b) under a gradient of cellular chiral torque (\( \lambda = 0.1 \)).](image)
IV. DEFINITION OF THE BOUNDARY LAYERS

We systematically defined the boundary layers near either the bottom or the top boundaries. We discretized the area into $N_y$ layers from the 0th layer to the $(N_y - 1)$th layer. The $i$th layer is defined as the layer ranging from $y = iL_y/N_y$ to $y = (i + 1)L_y/N_y$ ($i = 0 \sim (N_y - 1)$). The boundary layers are defined as the 0th~$N_b$th and $N_t$th~$(N_y - 1)$th layers, respectively for those near the bottom and top boundaries.

We calculated the average velocity of each layer by averaging the velocity of cells in the layer in time, and drew the velocity profiles as shown in Fig. S3(a) ($N_y = 40, \lambda = 0.01, P_0 = 3.33$). Using the velocity profile, we defined the boundary layers via the following procedure.

1. We calculated the approximate bulk velocity $V'_{\text{bulk}}$ and the approximate standard deviation $\sigma'_{\text{bulk}}$ of the bulk velocity by averaging the velocity of the 15th~24th layers.

2. We smoothed the velocity profile to obtain $v_i = v_{\text{sm}}(i)$ by a simple moving average, where the mean is taken from two data on either side of a central value.

3. We determined $N_b$ as the maximum integer $i < 15$ which satisfies either $v_{\text{sm}}(i) < V'_{\text{bulk}} - 3\sigma'_{\text{bulk}}$ or $v_{\text{sm}}(i) > V'_{\text{bulk}} + 3\sigma'_{\text{bulk}}$.

$N_t$ is determined as the minimum integer $i > 24$ which satisfies either $v_{\text{sm}}(i) < V'_{\text{bulk}} - 3\sigma'_{\text{bulk}}$ or $v_{\text{sm}}(i) > V'_{\text{bulk}} + 3\sigma'_{\text{bulk}}$.

In Fig. S3(a), we show the $N_b$th and $N_t$th layers, determined by the above procedure, with the square markers on examples of velocity profiles ($N_y = 40, \lambda = 0.01, P_0 = 3.33$).

Fig. S3. (a) Examples of the flow profiles are shown ($N_y = 40, \lambda = 0.01, p_0 = 3.33$). The $N_b$th and $N_t$th layers, determined by the above procedure, are also shown with the square markers. (b) Spatial profiles of shape index $\langle q_\alpha \rangle$ are shown. The $N_b$th and $N_t$th layers are also shown with the square markers. The data points for the top and bottom layers are marked with dashed ellipsoids as a guide for eye. In (a) and (b), each data point corresponds to each layer.

V. RELATIONSHIP BETWEEN $\langle q_\alpha \rangle_{\text{BL}}$ AND $\langle q_\alpha \rangle_{\text{BU}}$

In Fig. S3(b), we show examples of spatial profiles of the shape index $\langle q_\alpha \rangle$. We find that the shape index for the top and bottom layers are outlier due to the flat boundaries which induce large cell deformation. To avoid the outliers, we eliminated the top and bottom layers when we calculated $\langle q_\alpha \rangle_{\text{BL}}$.

In Fig. S4(a), we show the dependence of $\langle q_\alpha \rangle_{\text{BU}}$ on the target shape index $p_0$ for the data in Fig. 5 in the main text ($N_y = 40, \lambda = 0.01, P_0 = 3.33$). We found that larger $\sigma$ and $p_0$ provide larger $\langle q_\alpha \rangle_{\text{BU}}$.

In Fig. S4(b), we show the relationship between $\langle q_\alpha \rangle_{\text{BU}}$ and $\langle q_\alpha \rangle_{\text{BU}}$ for the same data set. We find $\langle q_\alpha \rangle_{\text{BU}} \approx \langle q_\alpha \rangle_{\text{BL}}$. As shown in Fig. S4(b), the spatial profiles of the shape index are nearly homogeneous except on the top and bottom layers. Hence, we obtain $\langle q_\alpha \rangle_{\text{BU}} \approx \langle q_\alpha \rangle_{\text{BL}}$. 
Fig. S4. (a) $\langle q_\alpha \rangle_{BU}$ is plotted against $P_0$ for different values of $\sigma$ for a large system size $N_y = 40$. The torque gradient $\lambda$ is fixed to 0.01. The different symbols are for three different samples. (b) $\langle q_\alpha \rangle_{BU}$ is plotted against $\langle q_\alpha \rangle_{BL}$ for different $\sigma$ and $P_0$. The dashed line indicates $\langle q_\alpha \rangle_{BU} = \langle q_\alpha \rangle_{BL}$. The different symbols are for three different samples.