Nanowire Magnetoscope Reveals a Cellular Torque with Left-Right Bias

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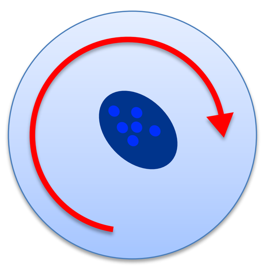
Introduction

Mechanical forces between cells and microenvironment regulate many physiological functions, for example, greater cell contractile force is correlated with the apoptosis, proliferation, and osteogenic differentiation. Clearly, the ability to characterize cellular force is crucially important.

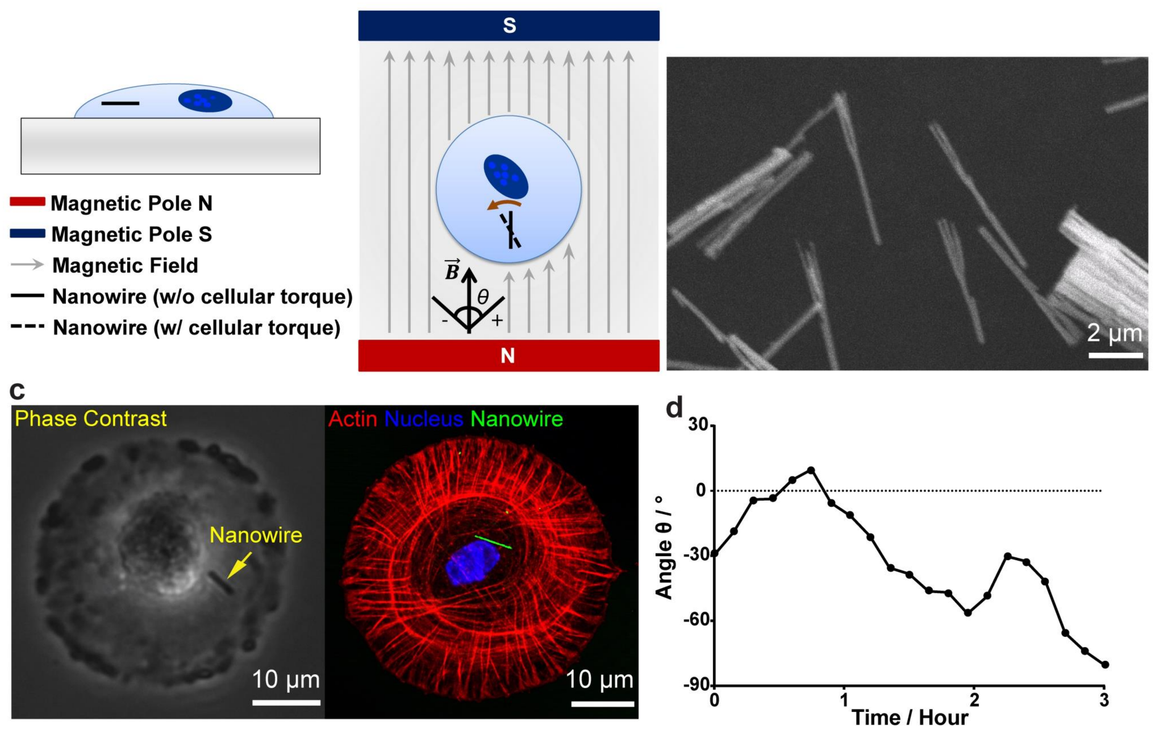
Recently, a variety of cell mechanics was reported with left-right (LR) asymmetry. Evidences were found cell alignment, migration, and cell-cell assembly. In particular, actomyosin activity was found essential for such LR asymmetric mechanics. Because actomyosin activity is also associated with cellular force, it predicts a type of cellular force, e.g. *rotational force*, that mediates the LR-biased mechanics and eventually coordinates the formation of LR asymmetry in tissue architecture. **However, Due to the lack of appropriate platform, cellular rotational force with LR bias has yet to be found.**



Rotational forces with LR bias ?

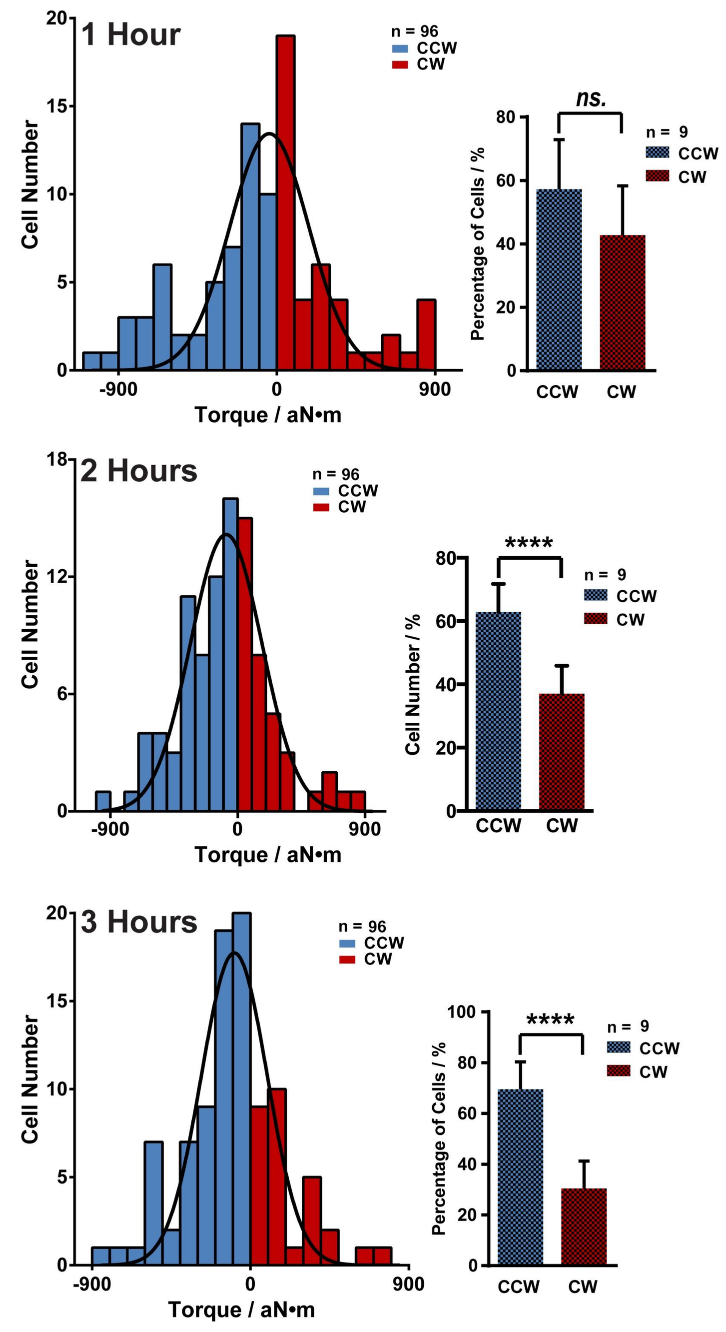
Approach

Nanowire magnetoscope reveals a rotating force – torque – generated by cells.



Working Principle: Ferromagnetic nanowires were deposited and internalized by micropatterned cells. Within a uniform, horizontal magnetic field, the nanowires that initially aligned with the magnetic field were subsequently rotated due to the cellular torque. Because the moment of inertia of nanowires is negligible, the rotational angle of each snapshot can be considered as the quasi-steady-state, where the magnetic moment and the cellular torque can be considered as in equilibrium. As such, the cellular torque was measured according to $\tau = \mu \cdot B \cdot \sin\theta$, where τ is the magnetic torque, μ is magnetic moment of nanowires, B is the external magnetic field, and θ is the rotational angle of nanowire.

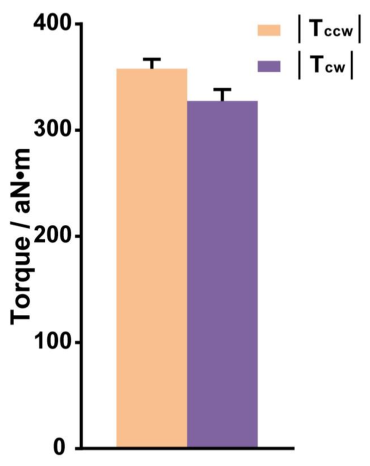
Result I: LR-Biased Cellular Torque



- Using Time-lapse microscopy tracing the cellular torque in a 3-hour duration, the torque representing each single cell was calculated based on the averaged torque measured within a single cell during the entire time course.

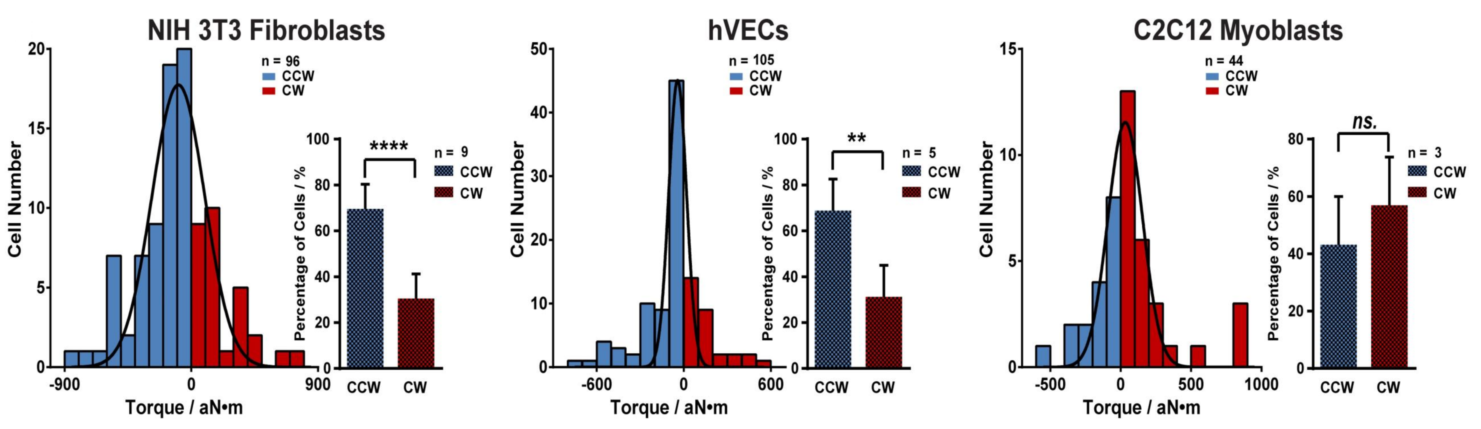
Cellular torque is LR-biased, i.e. CCW-biased torque by NIH 3T3 fibroblasts, and the bias increased over time.

The magnitude of the cellular torque: 300-350 aN·m for NIH 3T3 fibroblasts.

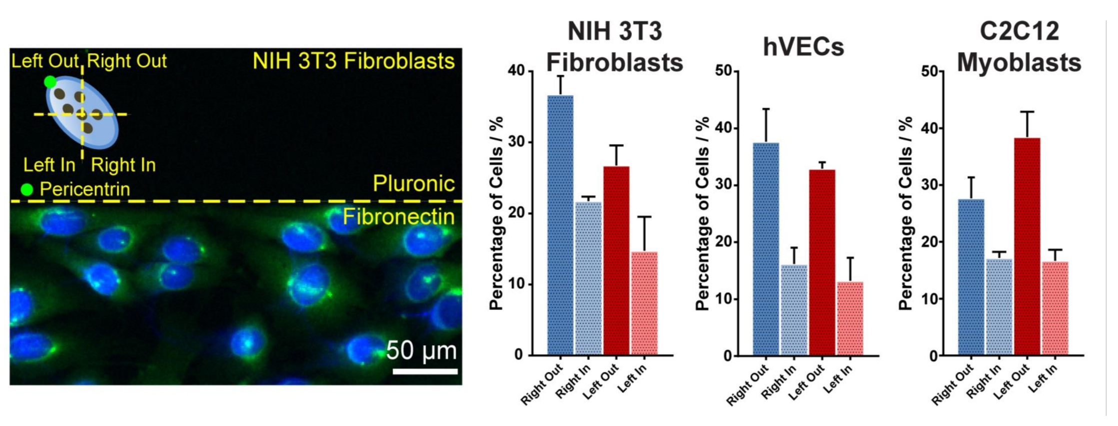


Result II: Cell-Type Dependence

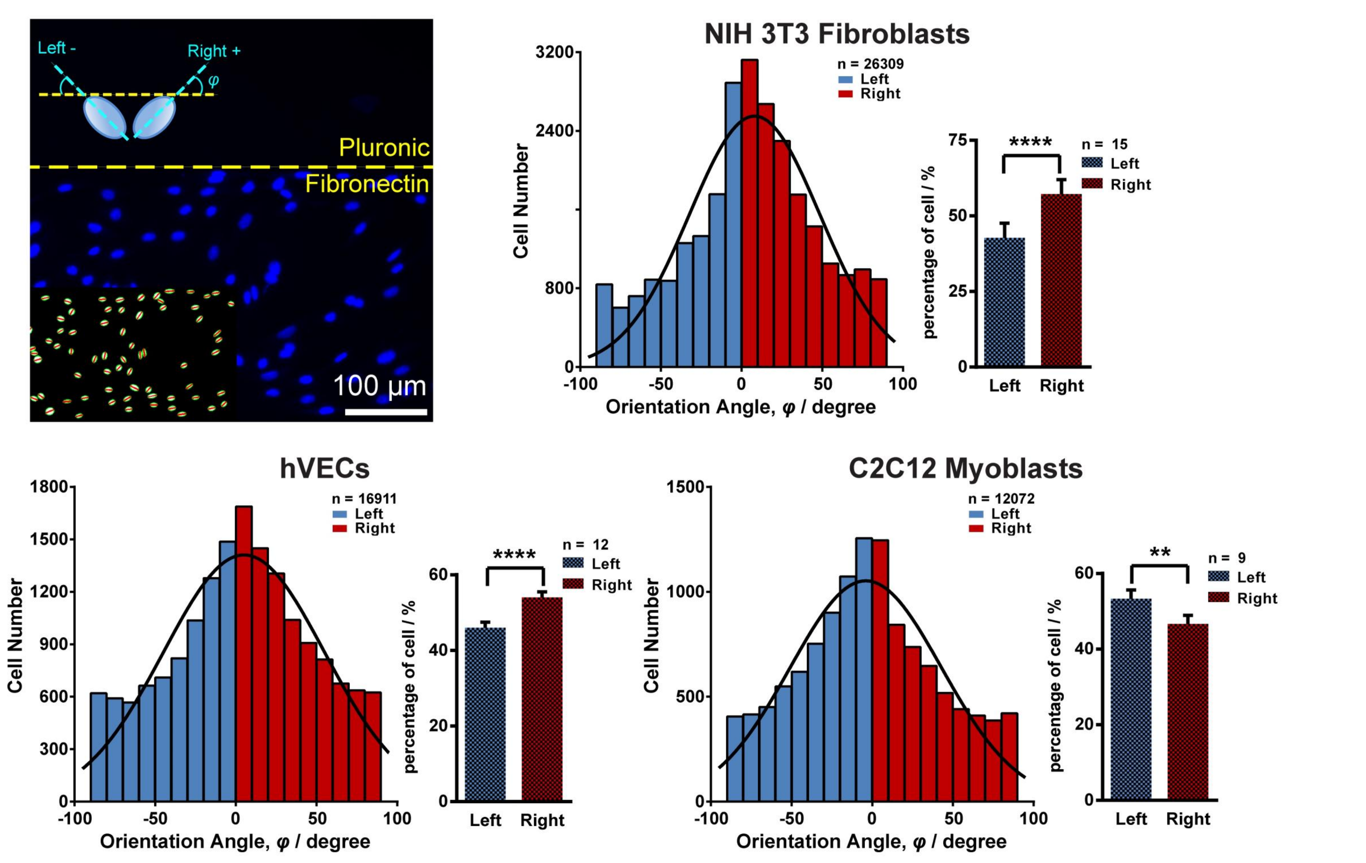
LR-biased cellular torque depending on cell types



LR-biased cell polarity depending on cell types



LR-biased cell orientation depending on cell types

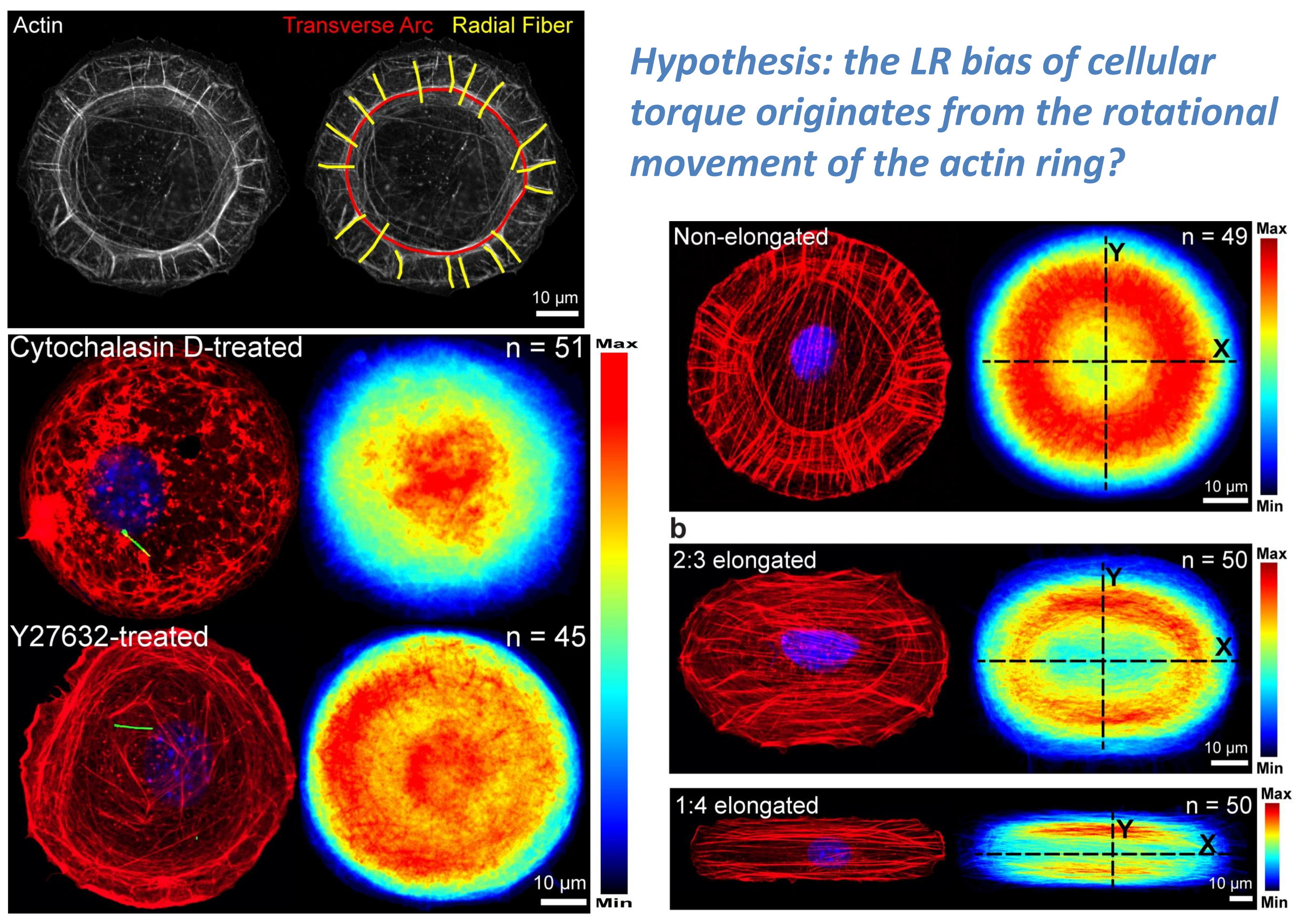


We further applied the nanowire magnetoscope for other adherent cells. Human vascular endothelial cells (hVECs) exhibited a CCW-biased torque, while C2C12 myoblasts showed a CW-biased torque. This cell type-dependence was also reported in other types of LR asymmetries, *i.e.* cell orientation and polarity, suggesting a possibility that other types of LR-biased cell behavior may be closely associated with this LR-biased torque. The result showed that NIH 3T3 fibroblasts, which exhibited CCW-biased torque, had a rightward bias in their polarity and orientation. Consistently, the CCW bias of hVECs and CW bias of C2C12 myoblasts also led to a rightward and leftward polarity/orientation, respectively. Together, it suggests that the LR-biased cellular torque is cell-type dependent and associated with other types of LR asymmetry.

Result III: Subcellular Actin Ring

Formation of a subcellular actin ring

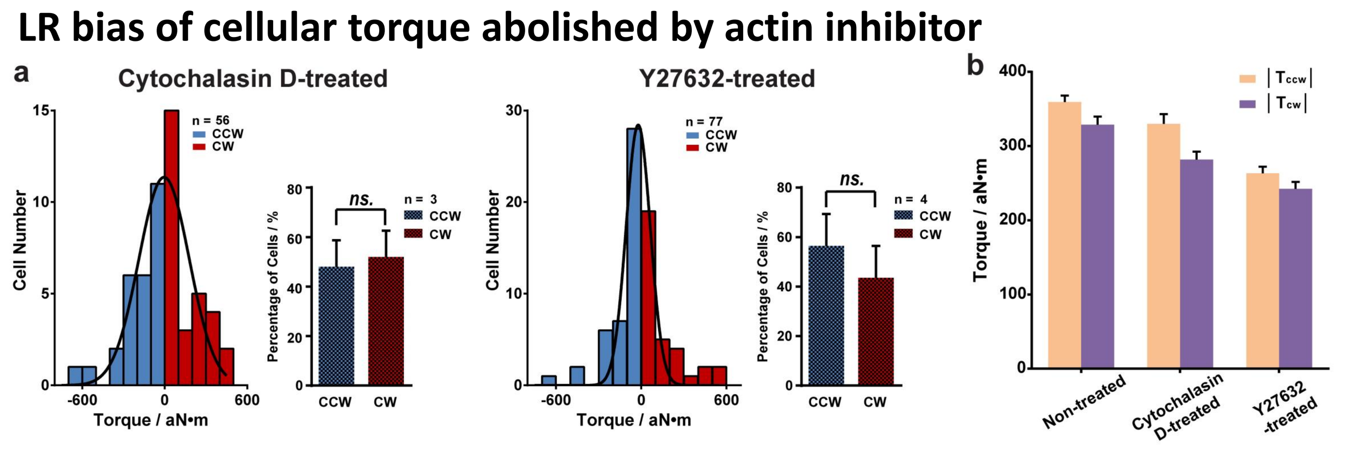
Hypothesis: the LR bias of cellular torque originates from the rotational movement of the actin ring?



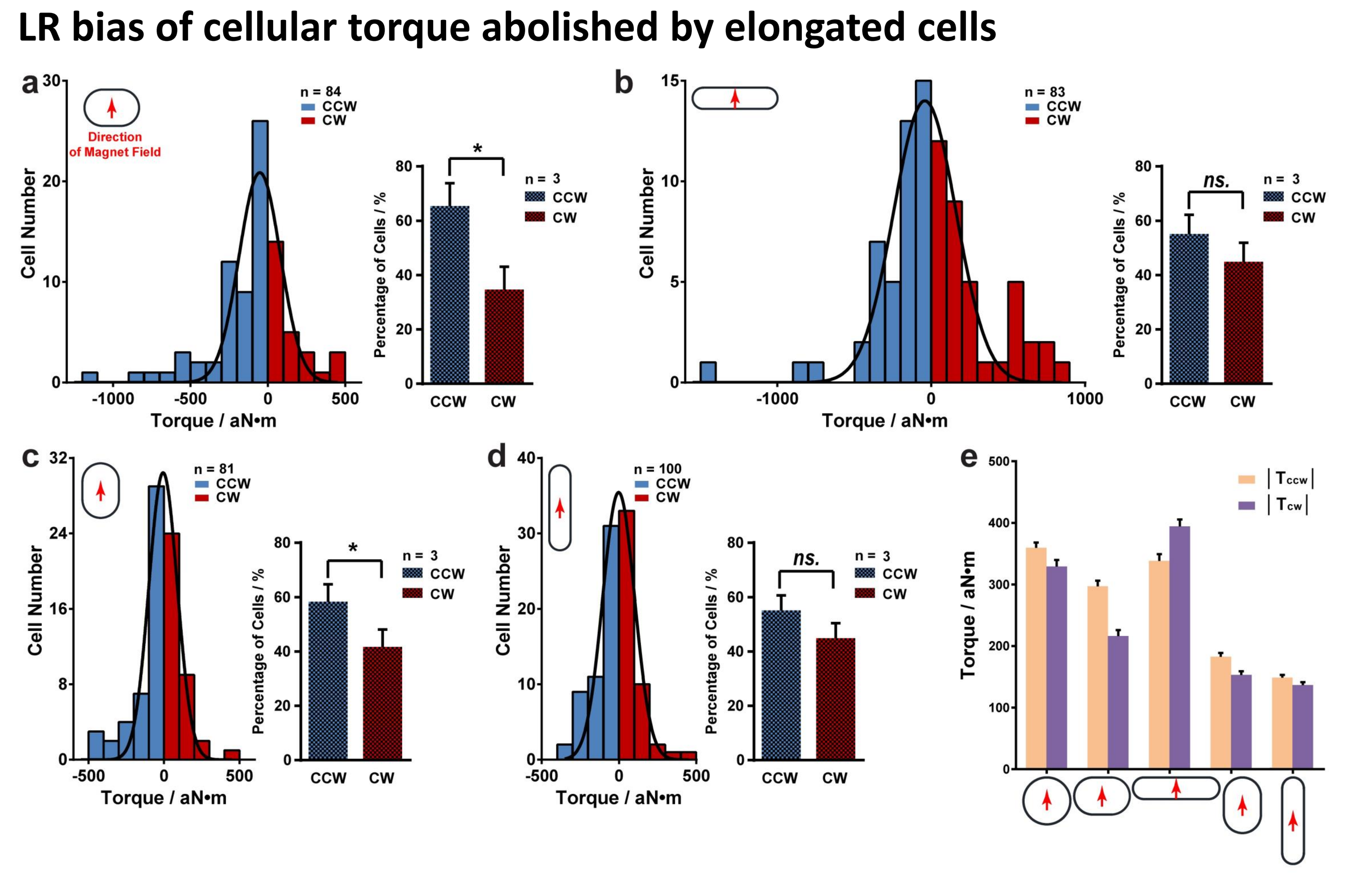
Inside the micropatterned cell with circular shape, there exists a ring structure composed of two classes of actin filaments: radial fibers and transverse arc. Transverse arcs are curved actin filaments in parallel with cell periphery, while radial fibers are actin filaments that intersect with transverse arc and point towards the periphery. After image stacking, there was a clear actin ring around the cell periphery.

We found that the actin ring is the key factor determining the LR bias of the cellular torque. Using two different approaches that disrupt different aspects of actin ring, *i.e.* disassembly of actin filaments by actin inhibitor, cytochalasin-D or Y27632, or disruption of actin ring by elongated cell shape, the LR bias was all abolished.

LR bias of cellular torque abolished by actin inhibitor



LR bias of cellular torque abolished by elongated cells



Conclusion

We report a nanowire magnetoscope that reveals a cellular torque based on internalized nanowires. This torque is LR-biased depending on cell types, and associated with other types of LR asymmetry. Moreover, we found that an actin ring composed of transverse arc and radial fibers is the key factor determining the LR bias of cellular torque. Our finding provides a fundamental origin of cytoskeletal chirality. More broadly, we anticipate that our method will provide a perspective on mechanics-related cell physiology and force transmission necessary for LR propagation in tissue formation.

References:

W. Liu, Y. Bao, M. L. Lam, T. Xu, K. Xie, H. S. Man, E. Y. Chan, N. Zhu, R. H. W. Lam, and T.-H. Chen, ACS Nano, 2016, accepted.*

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