A TOOL FOR STUDYING THE ROLE OF INTERCELLULAR STRESSES AND DYNAMIC CELL SHAPE MODULATIONS IN MECHANICAL INHIBITION OF CELL DIVISION

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INTRODUCTION

Cell migration is engaging in key physiological processes such as embryogenesis, wound healing, angiogenesis, and immune response. Unfortunately, it is also included in various pathologies, such as inflammation and cancer metastasis. In each of these physiological and pathological processes, cell migration involves the same basic mechanism of cell movement along or through tissue. This movement is driven by cellular forces, exerted through focal adhesions on the substrate or through cell-cell junctions with neighboring cells [1], and most importantly is also accompanied by extensive cell division events. It has been proposed that cell area [2] and cell-cell forces [3] impact various phases of the cell cycle during the expansion process of an epithelial layer. However, when it comes to cell cycle control during the more common physiological state of condensed epithelial tissues [4], the nature of size-based and force-based mechanisms and how they might be compiled to control the cell cycle transition and duration is still unclear. Here we map two-dimensional traction forces and intercellular stresses in highly dense epithelial tissue that still maintains unique dynamic patterns. We are now able to simultaneously track thousands of cells in a tissue, and track each cell; its morphology, progression in cell cycle state, and acting mechanical forces.

METHODS

As a model system for computing cellular forces [5], we prepared polyacrylamide gel as a substrate for a condensed colony of MDCK-Fucci cells (express Ctd1-red fluorescent protein during G1 and S phases and geminin green fluorescent protein during S-G2-M). We used traction force microscopy (TFM) to map traction force at the cell-substrate interface and monolayer stress microscopy (MSM) to map cell-cell stresses. We integrated our computational tools for stress measurement with image analysis tools, such as CellPose [6] and TrackMate [7].

RESULTS

We succeeded in accurately detecting the cell boundaries for highly dense epithelial tissue. We got an image analysis tool model that: 1. Skips gaps and extruded cells; 2. has an extremely high probability for successful segmentation; 3. Track cells in space and time, and relate spatiotemporal force patterns with individual cell morphology and cell cycle state.

CONCLUSION

The developed tool will become a cardinal pillar in our quest to decipher the complex relationship between the fundamental biological functionality of cell division, and physical properties such as force patterns, cell dynamics, and cellular morphology. But more so, our tool can be easily used for the study of mechanical effects on a variety of biological or molecular events such as differentiation, cell-cell communication via calcium signaling, and more.

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