CELL FORCE MEASUREMENT USING MEMS-BASED MICROPILLAR ARRAYS

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The mapping of cellular traction forces is crucial to understanding the means by which cells regulate their physiological function. Polymeric micropillar arrays have been used for measuring cellular traction force. However, the field-of-view of existing measurement methods is limited and the biocompatibility of micropillar arrays needs to be further improved. Since the submicron scale deflection of pillars is typically measured with a 60x objective lens, the field-of-view is limited, ensuring that only a few cells can be imaged at a time, thereby reducing the measurement efficiency.

To realize high-throughput cellular force mapping of multiple cells in a large field-of-view, a cell force measurement method was studied based on MEMS-based micropillar arrays. The force-displacement relationship of micropillar was achieved from analytic model and numerical simulation. The micropillar arrays were designed, fabricated and activated to yield force measurement resolution on the scale of nano newton and more in-vivo like microenvironment. The resulting PDMS micropillar was 2 μm in diameter and 6 μm in height.

For high throughput cell force mapping, double-sided micropillar arrays and the measuring system based on moiré effect were designed and validated. The double-sided micropillar arrays (DMA) integrate two independent micropillar arrays into a single sensor. Cells were seeded on the top side of the DMA, where they spread, contract and distort pillars under the cell body. The micropillars on the opposite side serve as a reference grating which enables the generation of a two-dimensional moiré pattern with the deflected pillars on the top side upon laser illumination through the DMA. When micropillars on the cell culture side are deflected due to cell contraction, the distorted moiré fringes are readily seen. The cells cultured on DMA may be analyzed using both conventional microscopy and moiré-based force mapping. Compared to the moiré-based method using two independent substrates, the use of DMA precludes the need for precise alignment of two independent samples, simplifying the system and improving moiré pattern contrast.

From the force map, it is clear that the maximum force was generated in the center of the cell, near the nucleus, with a force value of approximately 40 nN. Compared to the pillars in the center of the cell, the pillars under lamellipodia were not deflected, though some pillars were entirely involved in the cytoskeleton. The resulting force maps were consistent with the force mapping results using conventional microscopy image analyses, enabling the real-time acquisition of cell force maps from a larger field-of-view.

Using optical microscopy and confocal microscopy, cell force measurements were conducted and the image analysis method was discussed. Cell force of human hepatic stellate cell line (LX-2) in their activated status was quantified, which will aid disease study and drug screen related to liver fibrosis. Also the 3 days cell culture on the micropillar arrays indicates that the micropillar arrays was biocompatible.

Word Count: 479

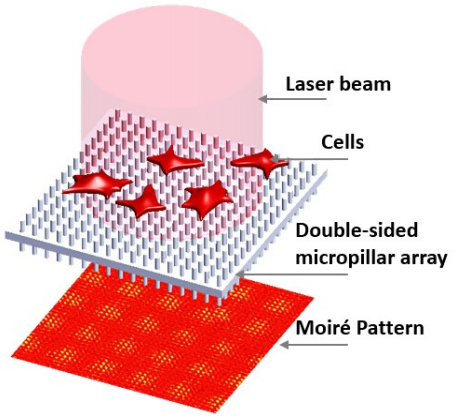

Fig. 4 Schematic of DMA: the top side of DMA is employed as the cell culturing substrate and the bottom side serves as reference grating for the formation of the two-dimensional moiré pattern.

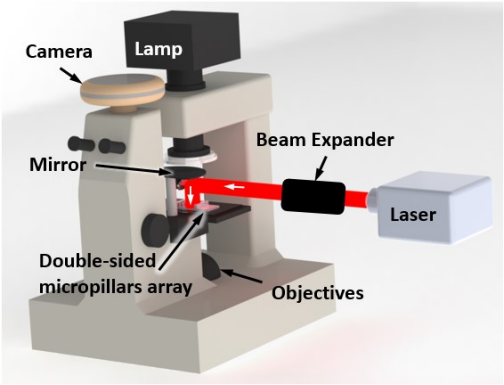

Fig. 2 System configurations for cell force mapping using DMA. The light source can be switched from laser to lamp by rotating the mirror in the light path, therefore both conventional microscopy and moiré-based force mapping can be conducted in the same system.

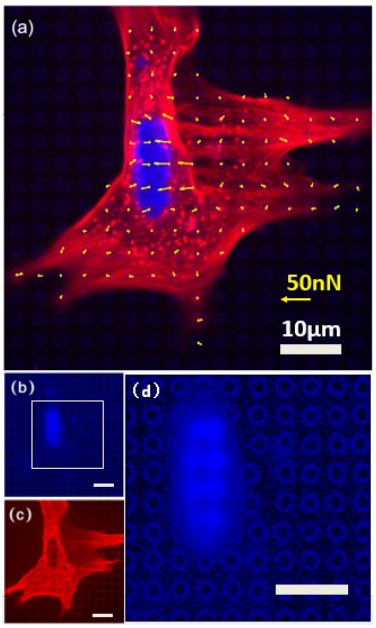

Fig. 3 NRVM force mapping using confocal microscopy (a) Confocal microscopic image of top surfaces of pillars. Blue: cell nucleus. Red: stress fiber consisting of actin. Yellow arrow: force vector. (b) (c) Images at the level of top surfaces of pillars from two fluorescent channels. Scale bar:10 μm (d) Enlarged to show the deflected pillars. Scale bar:10 μm

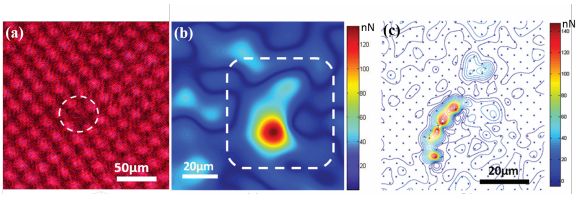

Fig. 4 (a) Moiré pattern in a larger field of view (20X objective); the circle indicates the distorted moiré grid. (b) Force map derived by moiré-based method. (c) Force map generated by calculating each pillar's displacement within the area of the dashed square in Fig. 4(b).

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