Fabricating Biomimetic Cilia to Study Viscous Fluid Flow

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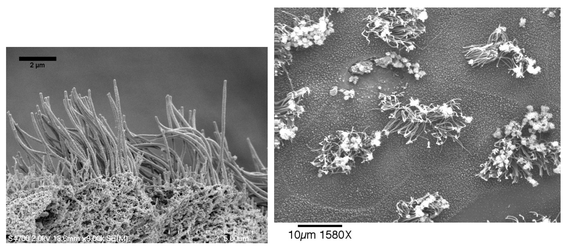
Abstract

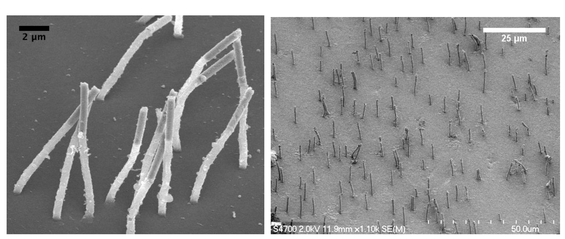
To understand fluid flow driven by cilia in biological systems such as in the lungs and in vertebrate embryos, we fabricated arrays of biomimetic cilia, core-shell microstructures that mimic the size of biological cilia. These biomimetic cilia are 10 microns tall and 600 nm in diameter with a flexible poly(dimethylsiloxane) (PDMS) core and an upper portion surrounded by a thin shell of nickel. To produce flow, they are immersed in a fluid and magnetically driven by a rotating permanent magnet. As cilia in biology are immersed in environments of varying viscosities and elasticities, biomimetic cilia have the potential to contribute to a deeper understanding of how fluids are moved at the micro-scale.

Artificial Cilia as Biological Mimics

Biological Cilia. Biological lung cilia are typically 7 μm tall and 250 nm in diameter. They grow in patches on cell surfaces in the lungs. Below are SEM images of human bronchial epithelial cells[1]. In vertebrate embryos, each cell grows a single cilium that is 5 μm tall and 300 nm in diameter.



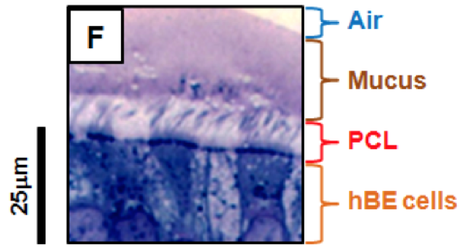
Biomimetic Cilia. Our cilia are 10 μm tall and 600 nm in diameter. They are created in arrays of 2 million cilia per square centimeter.



Cilia in Biological Systems

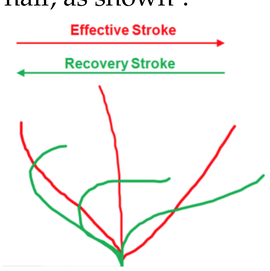
Cilia are a main driving force for liquids in many biological systems, such as in the lungs and in vertebrate embryos.

In the lungs. Cilia protrude from human bronchial epithelial (hBE) cells lining the airways and actively beat to transport mucus and pathogens out of the lungs, as shown in the histology image below[2].

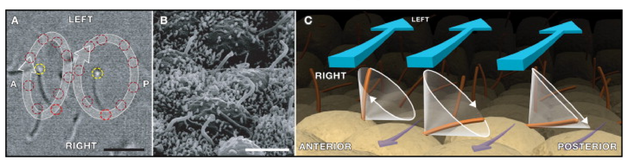


An airway cilium beats with a linear stroke, bending close to the cell surface for half its path and fully extending itself for the other half, as shown[2].

In cystic fibrosis, the mucus layer is so thick and viscous that beating cilia have difficulty transferring toxins in the mucus out of the airways. This inability to transport these materials can lead to chronic lung infections and ultimately death.



In vertebrate embryos. Nodal cilia are found in vertebrate embryos and play a role in determining the left and right sides of the embryo. They beat in a tilted conical shape, which creates a leftward directed flow as seen in the image below[3].



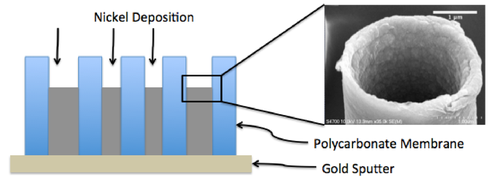
This leftward flow specifies the location of the heart and organs inside the body. In animals without nodal cilia, organ location is random[4].

References

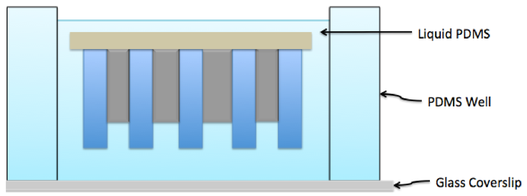
[1] Image at right taken by J. Carpenter (2008).
[2] Oldenburg, A., Chhetri, R. K., Hill, D., Button, B. (2012). Monitoring airway mucus flow and ciliary activity with optical coherence tomography. *Biomed. Opt. Express*, 3:1978–1992.
[3] Hirokawa, N., Tanaka, Y., Okada, Y., Takeda, S. Nodal Flow and the Generation of Left-Right Asymmetry. *Cell*, 125:33–45.
[4] Hirokawa, N., Okada, Y., Tanaka, Y. Fluid dynamic mechanism responsible for breaking the left-right symmetry of the human body: The nodal flow. *Annu. Rev. Fluid Mech.*, 41:53-72.

Artificial Cilia Fabrication

Nickel is deposited on a gold sputtered polycarbonate track-etched (PCTE) membrane. The gold layer is used for conduction to drive nickel into the pores of the membrane through an electrodeposition process. The Ni forms a tube as it deposits, as shown in the SEM at right.

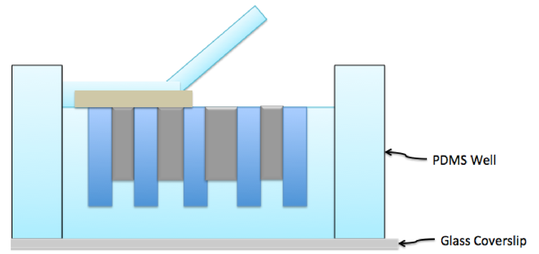


A 1 mm square of Ni tube-filled PCTE membrane is immersed in liquid PDMS such that the PDMS fills the Ni tubes and is then set into a well structure Au layer facing upward. The well structure is used as a fluid reservoir. The entire sample is heated overnight at 80°C to cross-link the liquid PDMS.

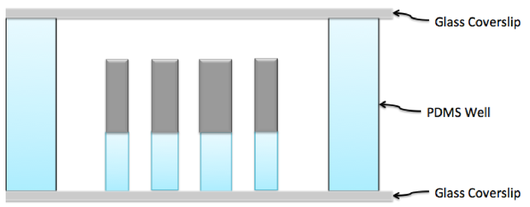


The cross-linked PDMS covering the Au layer is removed with tweezers. The Au is scraped off, leaving the PCTE membrane with the PDMS and Ni layer bare.

The entire well is immersed for 3 minutes in dichloromethane (DCM) at 60°C to dissolve the PCTE membrane and free the artificial cilia.



Contact between air and cilia is avoided, as air bubbles can cause the cilia to collapse. The well is rinsed in ethanol to remove DCM, and the cilia array is ready to be used. When the array is put on a microscope, a clean coverslip is placed on top of the well as a lid, preventing fluid evaporation during experiments.



Future Work: Fluid Flow with Artificial Cilia

Red one micron fluorescent microspheres will be added to multiple cilia arrays and videos will be taken at 30 frames/sec to trace the response of different fluids to beating cilia. The cilia beat is obtained by rotating a permanent magnet above the array. Below are minimum and maximum intensity tracks over 30 sec of beating biomimetic cilia (at left) and the resulting paths of microspheres (at right) 10 μm above the cilia tips in phosphate buffered saline. Future work will include tracking the microspheres in different viscosities of a purely viscous sucrose solution and eventually in viscoelastic solutions, such as mucus.