Determining the Mechanics of Living Cells by Atomic Force Microscopy

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The atomic force microscope (AFM) has become an important tool in the developing field of bionanotechnology. The AFM uses a sharp tip on the end of a cantilever to scan over a sample at a constant deflection to determine the topography of a surface. (Figure 1) The sample scanner moves in the x-y plane and is controlled by a piezoelectric crystal. Another piezoelectric crystal is coupled with a feedback loop to control the z-direction motion of the cantilever and keep the deflection of the tip against the surface constant. [1] Frequently used as an imaging tool, the AFM can provide detailed images of biologic surfaces, and has shown advantages over transmission electron microscopy because AFM can be used to monitor hydrated cells and even living cells. [2] In addition to imaging, the AFM can be very useful as a tool for nanoindentation and investigating surface forces of samples. Utilizing the strengths of the AFM, researchers are able to determine the mechanical properties of living cells.

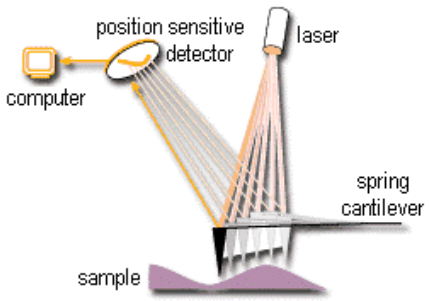

Figure 1: Basic Diagram of AFM.
http://www.che.utoledo.edu/nadarajah/webpages/whatsafm.html

In animals, nerve cells connect each part of the body with the brain. This neural network is a giant electric circuit that is essential. How this network forms is still a puzzle to scientists. In development, one cell can stretch a neural process from the spinal cord to the extremities. [3] Biologists have determined some of the chemistry behind this growth, but lack understanding of the basic mechanics of these nerve cells. The ultimate goal of this research project is to nanoindent a

growth cone of a living nerve cell from the sea slug *Aplysia californica* and to functionalize the AFM tip with anti-apCAM (antibodies which will bind to the membrane protein, apCAM, associated with the cytoskeleton of the cell) which will attach to the growth cone to monitor the forces the cell exerts on the tip. [4] (Figure 2) The difficulty of this experiment is that animal cells are very soft and delicate. In order to investigate the properties of living cells, much preparation and practice is needed to become adept operating the instrument.

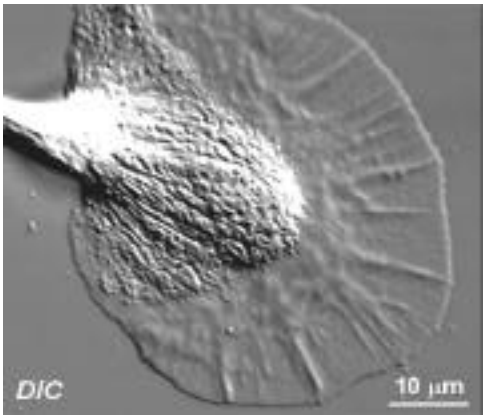

Figure 2: Growth Cone of Aplysia cell.
Courtesy of Prof. Daniel Suter, Purdue University [3]

To obtain practice indenting soft objects, the AFM is used to determine the elasticity of polydimethylsiloxane (PDMS), a polymer.[5] Next, living yeast cells are imaged and nanoindented while trapped in a porous membrane.[6] (Figure 3) Yeast cells have a cell wall which makes them more resistant to abuse from a developing researcher. The cell wall also prevents the yeast cells from strongly adhering to a surface. It helps the cell to maintain a defined shape and by doing so restricts the surface area in contact with the glass slide. The small forces produced by the AFM tip would sweep away the yeast cells on the surface leaving none to image or indent. The pores of the membrane will trap the cells and prevent them from being swept away.

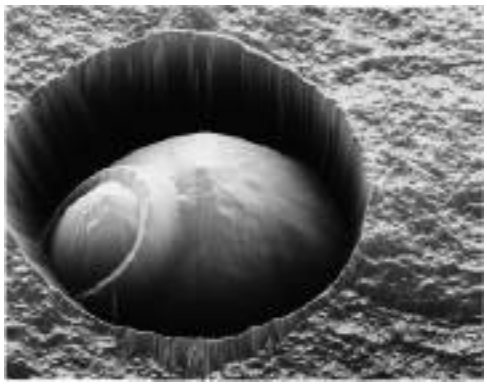

Figure 3: Yeast cell trapped in membrane. [6]

Once enough practice has been obtained, living nerve cells can be indented. Thus far, only PDMS has been indented. The remainder of this research will be continued in the fall.

The field of bionanotechnology is rapidly advancing and new discoveries of cell function are constantly being discovered. Discovering how a nerve cell manages to form a connection from the spinal cord to the pinky toe is an intriguing puzzle. This challenge requires the knowledge of the chemistry and mechanics involved. The mechanics of a living cell have never been completely determined, and the AFM shows promise in aiding researchers to determine these mechanical properties.

Sources

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