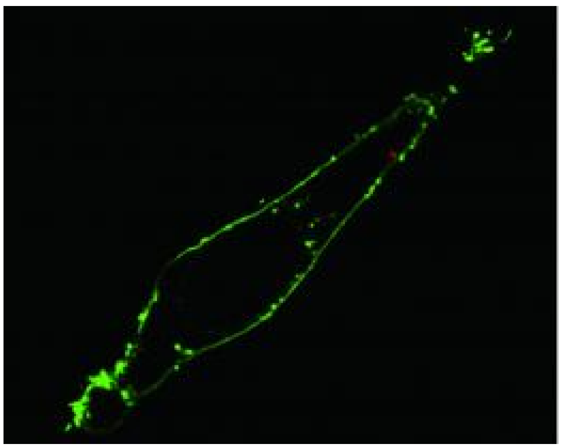
A single cell endoscope: Researchers use nanophotonics for optical look inside living cells

December 20 2011, by Lynn Yarris



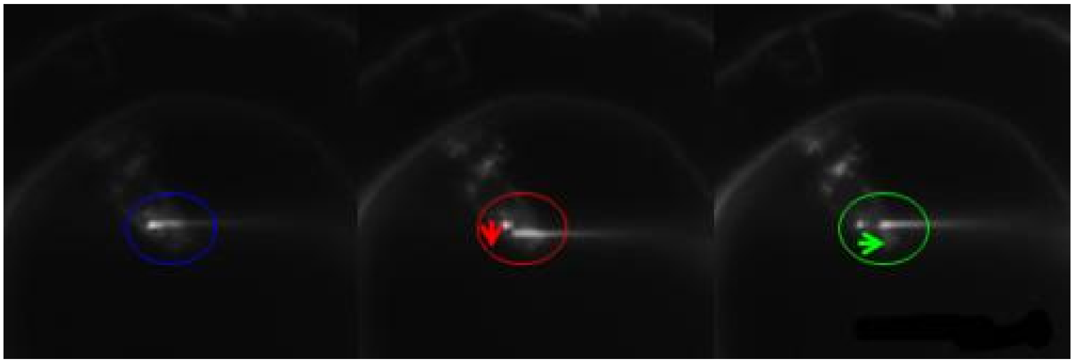
Fluorescence confocal image of a single living HeLa cell shows that via nanoendoscopy a quantum dot cluster (red dot) has been delivered to the cytoplasm within the membrane (green) of the cell. Courtesy of Berkeley Lab

(PhysOrg.com) -- An endoscope that can provide high-resolution optical images of the interior of a single living cell, or precisely deliver genes, proteins, therapeutic drugs or other cargo without injuring or damaging the cell, has been developed by researchers with the Lawrence Berkeley National Laboratory (Berkeley Lab). This highly versatile and mechanically robust nanowire-based optical probe can also be applied to biosensing and single-cell electrophysiology.

A team of researchers from Berkeley Lab and the University of California (UC) Berkeley attached a tin oxide nanowire waveguide to the tapered end of an optical fibre to create a novel endoscope system. Light travelling along the optical fibre can be effectively coupled into the nanowire where it is re-emitted into free space when it reaches the tip. The nanowire tip is extremely flexible due to its small size and high aspect ratio, yet can endure repeated bending and buckling so that it can be used multiple times.

"By combining the advantages of nanowire waveguides and fibre-optic fluorescence imaging, we can manipulate light at the nanoscale inside living cells for studying biological processes within single living cells with high spatial and temporal resolution," says Peidong Yang, a chemist with Berkeley Lab's Materials Sciences Division, who led this research. "We've shown that our nanowire-based endoscope can also detect optical signals from subcellular regions and, through light-activated mechanisms, can deliver payloads into cells with spatial and temporal specificity."

Yang, who also holds appointments with the University of California Berkeley's Chemistry Department and Department of Materials Science and Engineering, is the corresponding author of a paper in the journal *Nature Nanotechnology* describing this work titled "Nanowire-based single-cell endoscopy." Co-authoring the paper were Ruoxue Yan, Ji-Ho Park, Yeonho Choi, Chul-Joon Heo, Seung-Man Yang and Luke Lee.



Images of a nanowire endoscope in close contact with a quantum dot cluster in a HeLa cell (left), and separated vertically from the cluster by 2 mm (middle) and horizontally by 6 mm (right). Colored circles and arrows mark the position of the cluster and movement of the endoscope.

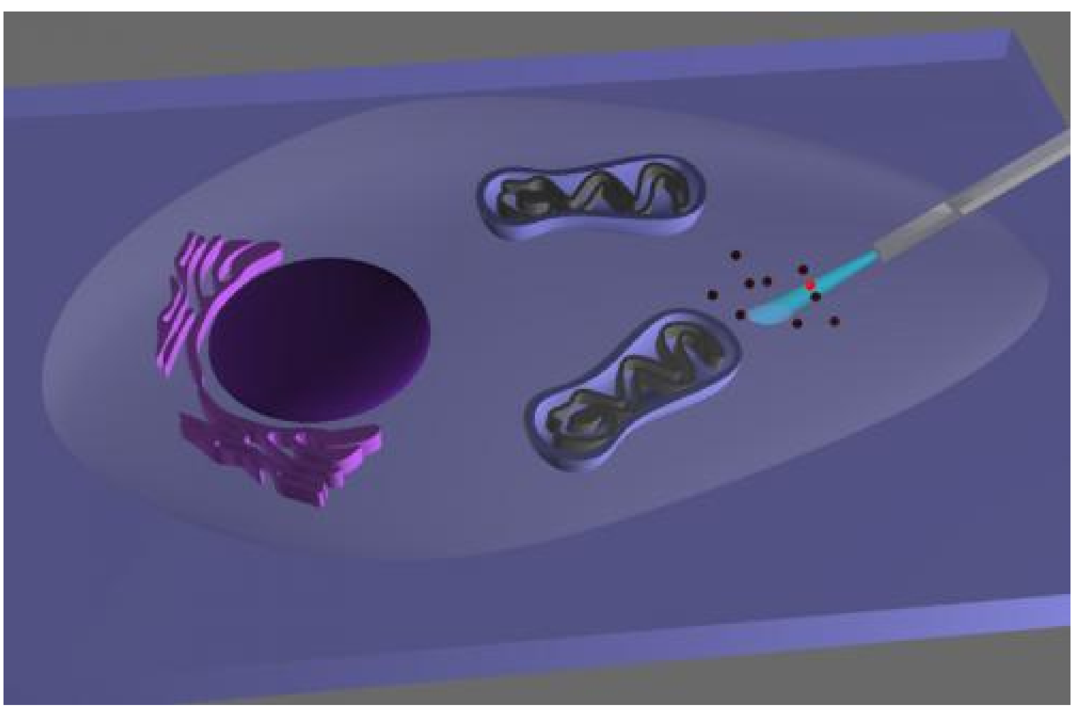
Despite significant advancements in electron and scanning probe microscopy, visible light microscopy remains the workhorse for the study of biological cells. Because cells are optically transparent, they can be noninvasively imaged with visible light in three-dimensions. Also, visible light allows the fluorescent tagging and detection of cellular constituents, such as proteins, nucleic acids and lipids. The one drawback to visible light imaging in biology has been the diffraction barrier, which prevents visible light from resolving structures smaller than half the wavelength of the incident light. Recent breakthroughs in nanophotonics have made it possible to overcome this barrier and bring subcellular components into view with optical imaging systems. However, such systems are complex, expensive and, oddly enough, bulky in size.

"Previously, we had shown that subwavelength dielectric nanowire waveguides can efficiently shuttle ultraviolet and visible light in air and fluidic media," Yang says. "By incorporating one of our nanophotonic components into a simple, low-cost, bench-top fibre-optical set-up, we were able to miniaturize our endoscopic system."

To test their nanowire endoscope as a local light source for subcellular imaging, Yang and his co-authors optically coupled it to an excitation laser then waveguided blue light across the membrane and into the interiors of individual HeLa cells, the most commonly used immortalized human cell line for scientific research.

"The optical output from the endoscope emission was closely confined to the nanowire tip and thereby offered highly directional and localized illumination," Yang says. "The insertion of our tin oxide nanowire into the cell cytoplasm

did not induce cell death, apoptosis, significant cellular stress, or membrane rupture. Moreover, illuminating the intracellular environment of HeLa cells with blue light using the nanoprobe did not harm the cells because the illumination volume was so small, down to the picolitre-scale."



This schematic depicts the subcellular imaging of quantum dots in a living cell using a nanowire endoscope. Credit: (Courtesy of Berkeley Lab)

Having demonstrated the biocompatibility of their nanowire endoscope, Yang and his co-authors next tested its capabilities for delivering payloads to specific sites inside a cell. While carbon and boron nitride nanotube-based single-cell delivery systems have been reported, these systems suffer from delivery times that range from 20-to-30 minutes, plus a lack of temporal control over the delivery process. To overcome these limitations, Yang and his co-authors attached quantum dots to the tin oxide nanowire tip of their endoscope using photo-activated linkers that can be cleaved by low-power ultraviolet radiation. Within one minute, their functionalized nanowire endoscope was able to release its quantum dot cargo into the targeted intracellular sites.

"Confocal microscopy scanning of the cell confirmed that the quantum dots were successfully delivered past the fluorescently labeled membrane and into the cytoplasm," Yang says. "Photoactivation to release the dots had no significant effect on cell viability."

The highly directional blue laser light was used to excite one of two quantum dot clusters that were located only two micrometers apart. With the tight illumination area and small separation between the light source and the dots, low background fluorescence and high imaging contrast were ensured.

"In the future, in addition to optical imaging and cargo delivery, we could also use this nanowire endoscope to electrically or optically stimulate a living cell," Yang says.

The nanowires used in these experiments were originally developed to study size-dependent novel electronic and optical properties for energy applications.

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