Video Article

August 2012: This Month in JoVE

Wendy Chao[1], Aaron Kolski-Andreaco[2]

[1]Department of Ophthalmology, Massachusetts Eye and Ear

[2]JoVE Content Production

Correspondence to: Aaron Kolski-Andreaco at aaron.kolski-andreaco@jove.com

URL: https://www.jove.com/video/5016
DOI: doi:10.3791/5016

Keywords: This Month in JoVE, Issue 66

Date Published: 8/1/2012

Citation: Chao, W., Kolski-Andreaco, A. August 2012: This Month in JoVE. *J. Vis. Exp.* (66), e5016, doi:10.3791/5016 (2012).

Abstract

Traditional microscopy requires lens objectives to magnify specimens, and can involve numerous optical components like additional objectives, filters, and mirrors to refract and direct light to optical sensors. The August 2012 issue of *JoVE (Journal of Visualized Experiments)* is marked by the third publication from the Ozcan Lab (University of California, Los Angeles) on their lens free lens-free "on-chip" microscopy platform, which they have pioneered.

Back in 2009, the Ozcan lab published their first article, which demonstrated the use of a lens-free imaging platform that could image cells within whole blood. This imaging technique involves placing a sample directly on an optoelectric sensor array, like the CCD or CMOS chips found in the standard camcorder. Then, LED illumination from above the sample casts shadows on the chip, which are recorded as digital holograms that can be subjected to image processing. Not only does this lensless imaging system allow for an extremely large field of view (FOV of 0.6-8 cm^2), the lack of bulky optical components makes it excellent for microfluidic applications. The Ozcan group demonstrates this in their follow-up 2011 JoVE article, in which fluorescently labeled white blood cells are imaged within a microfluidic chip.

This August in JoVE, the Ozcan group demonstrates further advancements to their lens-free microscopy platform by applying it to tomographic imaging of the nematode, *C. elegans*. By changing the angle of illumination, using a motorized rotation stage, the Ozcan group shows how multiple holographic slices may be generated for a sample-which, following processing, can be combined into tomograms. The Ozcan group confirms that their 3D reconstruction of the worm is successful by citing that the pharyngeal tube, a 5-micron-thick structure, is only visible in a single slice and in a 3D reconstructed cross section.

The compact and inexpensive nature of the lens-free imaging systems - designed and assembled by the Ozcan group - make them well suited for point-of-care diagnostic applications, or even cell phone based microscopy that can be performed in the field.

At the Ernest Gallo Clinic and Research Center at University of California, San Francisco JoVE learns how to visualize morphological changes in single cells using three-dimensional fluorescence microscopy and computer analysis. In this procedure, cells are given agonist, antagonist plus agonist, or no treatment, and prepared for immunofluorescence microscopy. Three-dimensional images are acquired with the help of a two-photon laser system, and the fluorescence data are processed on the computer using Imaris and Matlab.

To demonstrate the power of this technique, our authors quantify single-cell phenotypic changes in transfected HEK293 cells. Three-dimensional morphometric analysis shows dramatic changes in cells treated with agonist. These changes are not always apparent when using conventional 2D analysis. This technique allows morphological changes in single cells to be quantified and characterized in response to pharmacologic stimuli, offering a powerful new assessment tool to the arsenal of drug discovery research.

This month, in JoVE Applied Physics, we travel to South Carolina to visit the School of Materials Science and Engineering at Clemson University. Here, researchers show us how to construct lithium batteries for laboratory research.

The basic components of a lithium coin cell are the working electrode, the lithium electrolyte, and a lithium foil counter electrode. The different components of the coin cell are carefully assembled and sealed with a crimping machine. The finished coin cell can then be characterized in a battery tester. Using their homemade coin cells, our researchers can experiment with using different materials to make the cathodes of lithium coin cells.

This month in JoVE Immunology and Infection, we head to Basel, Switzerland to witness two techniques for diagnosing helminth infections. We also see how investigators perform the shuttle run test in children, in order to assess the effects of these infections on physical fitness.

Helminths are parasitic worms that can live inside humans and animals. Many species infect the intestinal tract, and may be transmitted through soil. Our authors demonstrate the Kato-Katz technique, which is used for diagnosing several common soil-transmitted helminths. They also demonstrate the Baermann technique, which uses light to stimulate the migration of certain types of phototactic helminths out of sample preparations. The various types of helminth species can subsequently be determined based on morphology.

Our authors then demonstrate how to set up and perform a 20-meter shuttle run test for children, and to measure the impact of helminth infections on physical fitness. This method can help determine the true burden of soil-transmitted helminth infections on overall health.

Copyright © 2012 Journal of Visualized Experiments

This brief summary highlights but a few notable video-articles that will be released in August. We'll also feature articles that demonstrate how to express genes in the midbrain of the chick embryo, study neutrophil transmigration in parallel flow chambers lined with endothelial cells, and precisely measure proteolytic degradation of the extracellular matrix.

Video Link

The video component of this article can be found at https://www.jove.com/video/5016/

Protocol

Determining Soil-transmitted Helminth Infection Status and Physical Fitness of School-aged Children

Peiling Yap[1, 2], Thomas Fürst[1, 2], Ivan Müller[1, 2], Susi Kriemler[1, 2], Jürg Utzinger[1, 2], Peter Steinmann[1, 2]
[1]Department of Epidemiology and Public Health, Swiss Tropical and Public Health Institute, Basel, Switzerland, [2]University of Basel, Basel, Switzerland

Chronic infection with soil-transmitted helminths (STHs) causes malabsorption, stunting, and wasting in the growing child. Hence, it is plausible that these infections also reduce the physical fitness of children. Here, we visualize two techniques for the diagnosis of STHs and the 20-meter shuttle run test for assessing children's physical fitness.

In ovo Electroporation in Chick Midbrain for Studying Gene Function in Dopaminergic Neuron Development

Ben Yang[1,*], Lauren B. Geary[1,*], Yong-Chao Ma[1, 2]
[1]Northwestern University Feinberg School of Medicine, Children's Hospital of Chicago Research Center, [2]Departments of Pediatrics, Neurology and Physiology, Northwestern University Feinberg School of Medicine
[*] These authors contributed equally

To assess the function and the regulation of genes during the development of midbrain dopaminergic neurons, we describe a method that involves *in ovo* electroporation of plasmid DNA constructs into embryonic chick ventral midbrain dopaminergic neuron progenitors. This technique can be used to achieve efficient expression of genes of interest to study different aspects of midbrain development and dopaminergic neuron differentiation.

Isolation of Human Umbilical Vein Endothelial Cells and Their Use in the Study of Neutrophil Transmigration Under Flow Conditions

Anutosh Ganguly, Hong Zhang, Ritu Sharma, Sean Parsons, Kamala D. Patel
Department of Physiology and Pharmacology, University of Calgary

This article first describes a procedure for isolating human endothelial cells from umbilical veins and then shows how to use these cells to examine neutrophil transmigration under flow conditions. By using a low-volume flow chamber made from a polymer with the optical characteristics of glass, live-cell fluorescent imaging of rare cell populations is also possible.

Construction and Testing of Coin Cells of Lithium Ion Batteries

Archana Kayyar[1], Jiajia Huang[1], Mojtaba Samiee[1], Jian Luo[1, 2]
[1]School of Materials Science and Engineering, Clemson University, [2]Center for Optical Materials Science and Engineering Technologies, Clemson University

A protocol to construct and test coin cells of lithium ion batteries is described. The specific procedures of making a working electrode, preparing a counter electrode, assembling a cell inside a glovebox and testing the cell are presented.

Quantitative Measurement of Invadopodia-mediated Extracellular Matrix Proteolysis in Single and Multicellular Contexts

Karen H. Martin, Karen E. Hayes, Elyse L. Walk, Amanda Gatesman Ammer, Steven M. Markwell, Scott A. Weed
Department of Neurobiology and Anatomy, Program in Cancer Cell Biology, Mary Babb Randolph Cancer Center, West Virginia University

We describe the prototypical method for producing microscope coverslips coated with fluorescent gelatin for visualizing invadopodia-mediated matrix degradation. Computational techniques using available software are presented for quantifying the resultant levels of matrix proteolysis by single cells within a mixed population and for multicellular groups encompassing entire microscopic fields.

Lensfree On-chip Tomographic Microscopy employing Multi-Angle Illumination and Pixel Super-Resolution

Serhan O. Isikman[1], Waheb Bishara[1], Aydogan Ozcan[1, 2, 3]
[1]Electrical Engineering Department, University of California, Los Angeles , [2]Bioengineering Department, University of California, Los Angeles , [3]California NanoSystems Institute, University of California, Los Angeles

Lensfree optical tomography is a three-dimensional microscopy technique that offers a spatial resolution of <1 μm × <1 μm × <3 μm in x, y and z dimensions, respectively, over a large imaging-volume of 15-100 mm^3, which can be particularly useful for integration with lab-on-a-chip platforms.

Copyright © 2012 Journal of Visualized Experiments

An Analytical Tool that Quantifies Cellular Morphology Changes from Three-dimensional Fluorescence Images

Carolina L. Haass-Koffler[1, 2], Mohammad Naeemuddin[1], Selena E. Bartlett[1, 3]
[1]Medications Development, Ernest Gallo Clinic and Research Center, University of California, San Francisco , [2]Clinical Pharmacology and Experimental Therapeutics, University of California, San Francisco , [3]Translational Research Institute and the Institute for Health and Biomedical Innovation, Queensland University of Technology, Brisbane, Australia

We developed a software platform that utilizes Imaris Neuroscience, ImarisXT and MATLAB to measure the changes in morphology of an undefined shape taken from three-dimensional confocal fluorescence of single cells. This novel approach can be used to quantify changes in cell shape following receptor activation and therefore represents a possible additional tool for drug discovery.

Lensless On-chip Imaging of Cells Provides a New Tool for High-throughput Cell-Biology and Medical Diagnostics

Onur Mudanyali[1], Anthony Erlinger[1], Sungkyu Seo[1], Ting-Wei Su[1], Derek Tseng[1], Aydogan Ozcan[1, 2]
[1]Electrical Engineering Department, University of California, Los Angeles, [2]California NanoSystems Institute, University of California, Los Angeles

Lensfree on-chip imaging and characterization of cells is illustrated. This on-chip cell imaging approach provides a compact and cost-effective tool for medical diagnostics and high-throughput cell biology applications, making it especially suitable for resource poor settings.

Lensless Fluorescent Microscopy on a Chip

Ahmet F. Coskun, Ting-Wei Su, Ikbal Sencan, Aydogan Ozcan
Department of Electrical Engineering, University of California, Los Angeles

A lensless on-chip fluorescent microscopy platform is demonstrated that can image fluorescent objects over an ultra-wide field-of-view of e.g., >0.6-8 cm2 with <4μm resolution using a compressive sampling based decoding algorithm. Such a compact and wide-field fluorescent on-chip imaging modality could be valuable for high-throughput cytometry, rare-cell research and microarray-analysis.

Disclosures

No conflicts of interest declared.

Copyright © 2012 Journal of Visualized Experiments