

Video Article

## June 2012: This Month in JoVE

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### Abstract

Back in 1905, in what is now the Czech Republic, Eduard Zirm performed the first corneal transplantation surgery (keratoplasty), which restored vision to a patient blinded by corneal injury. Today, eye banks all over the world prepare, store, and distribute donated corneas to hospitals so that thousands of sight-saving keratoplasties can be performed every year. In June 2012, JoVE has its eye on two research groups, one from Italy and the other from Michigan, who demonstrate two distinct methods for corneal graft preparation prior to transplantation.

Our authors from Italy show us their technique for excising the cornea from the ocular globe, which involves first dunking it in a series of sterilizing solutions preparing it for sterile handling, creating an incision on the scleral surface, and ultimately separating the corneal-scleral rim away from the globe. Once the cornea is removed, the endothelial cell density and viability is checked and it is prepared for long-term storage, during which time the media is periodically tested in high-throughput fashion with other grafts deposited at the eye bank.

In corneal transplantation surgeries, full-thickness grafts are used in a procedure called penetrating keratoplasty; however, JoVE learns that in some diseased corneas, only the endothelial layer is affected. Our authors from the University of Michigan & Midwest Eye Bank demonstrate further processing of donor corneas with Descemet's stripping automated endothelial keratoplasty (DSAEK), a procedure that involves grafting only the corneal endothelial layer. This relatively recent procedure is made possible via the use of a microkeratome, a device that can precisely cut through the cornea so that the endothelial layer can be separated for transplantation. The uniformity and quality of the donor tissue is verified using slit lamp and specular microscopy, and the graft is stored for corneal transplantation surgery.

Together, two groups of authors, from different continents, have documented the steps for removing the entire cornea from the eye and isolating the posterior layer for endothelial keratoplasty - a process that decreases the risk of infection or graft rejection and improves a patient's ability to see.

In JoVE Neuroscience, a multidisciplinary team of clinicians and scientists introduce new research applications of the electrocorticography (ECoG). The ECoG is an invasive procedure that uses strip and grid electrodes placed directly on the brain to localize seizure foci in epilepsy patients. This procedure can also be used to map cortical regions that need to be spared during resective surgery. Cortical mapping is accomplished by monitoring whether stimulation of a given electrode results in the disruption of movement or speech. Patients are typically subjected to intracranial monitoring to locate seizure foci for about a week, and this duration provides a unique window of opportunity for researchers to study the human brain in action with the ECoG, which has better signal-to-noise properties and less susceptibility to recording artifacts than the noninvasive electroencephalography (EEG).

After determining baseline activity at rest, our authors record brain activity in the high gamma frequency range, during simple cognitive, or motor tasks. Then, these investigators demonstrate how to use the SIGFRIED software system to perform rapid, real-time functional mapping based on ECoG signals, which are further analyzed to provide information regarding the brain regions associated with particular tasks.

In Bioengineering, JoVE encounters a team of scientists who show that the microvasculature can be approximately recreated on a chip. This "chip" is actually a device with microfluidic channels that are coated with endothelial cells. SU-8 photolithography is used to etch the channel pattern onto a silicon wafer, which acts as a mold for PDMS. Once the device has been fabricated, it is seeded with endothelial cells, which are cultured in the channels. Thanks to the transparency of PDMS, the cells can be imaged via microscopy. This *in vitro* model of the microvasculature provides a controlled microenvironment that can be used to study normal hemodynamic processes as well as in hematologic disease. This short summary is a mere glimpse at some of JoVE's content for the month of June. Further investigation might lead one to methods for *in situ* hybridization of adult mosquito tissue and embryo, visualizing mitosis in drosophila, and differentiating embryonic stems cells into motor neurons. Stay tuned.

### Video Link

The video component of this article can be found at <https://www.jove.com/video/4467/>

### Protocol

#### Hybridization *in situ* of salivary glands, ovaries and embryos of vector mosquitoes

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Temporal and spatial gene expression analyses have a crucial role in functional genomics. Whole-mount hybridization *in situ* is useful for determining the localization of transcripts within tissues and subcellular compartments. Here we outline a hybridization *in situ* protocol with modifications for specific target tissues in mosquitoes.

## Studying Mitotic Checkpoint by Illustrating Dynamic Kinetochore Protein Behavior and Chromosome Motion in Living *Drosophila* Syncytial Embryos

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The kinetochore is where the SAC initiates its signal monitoring the mitotic segregation of the sister chromatids. A method is described to visualize the recruitment and turnover of one of the kinetochore proteins and its coordination with the chromosome motion in *Drosophila* embryos using a Leica laser scanning confocal system.

## A simplified technique for In situ excision of cornea and evisceration of retinal tissue from human ocular globe

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The paper describes a simplified technique to excise corneal and to eviscerate retinal tissues from the ocular globe of human cadaveric donors. The technique described here will help to excise good quality tissues to be used for transplantation, surgical or research purposes without damaging other tissues of the ocular globe.

## Efficient differentiation of mouse embryonic stem cells into motor neurons

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We developed a new protocol to improve efficiency of *in vitro* differentiation of mouse embryonic stem cells into motor neurons. The differentiated ES cells acquired motor neurons features as evidenced by expression of neuronal and motor neuron markers using immunohistochemical techniques.

## Corneal Donor Tissue Preparation for Endothelial Keratoplasty

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Endothelial corneal transplantation is a surgical technique for treatment of posterior corneal diseases. Mechanical microkeratome dissection to prepare tissue results in thinner, more symmetric grafts with less endothelial cell loss and improved outcomes. Dissections can be performed at the eye bank prior to corneal transplantation surgery.

## Endothelialized microfluidics for studying microvascular interactions in hematologic diseases

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A method to culture an endothelial cell monolayer throughout the entire inner 3D surface of a microfluidic device with microvascular-sized channels (<30 μm) is described. This *in vitro* microvasculature model enables the study of biophysical interactions between blood cells, endothelial cells, and soluble factors in hematologic diseases.

## Recording Human Electroencephalographic (EEG) Signals for Neuroscientific Research and Real-time Functional Cortical Mapping

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We present a method for collecting electrocorticographic signals for research purposes from humans who are undergoing invasive epilepsy monitoring. We show how to use the BCI2000 software platform for data collection, signal processing and stimulus presentation. Specifically, we demonstrate SIGFRIED, a BCI2000-based tool for real-time functional brain mapping.

## Disclosures

No conflicts of interest declared.