



Three-Dimensional In Vivo Imaging Of Blood Vessels

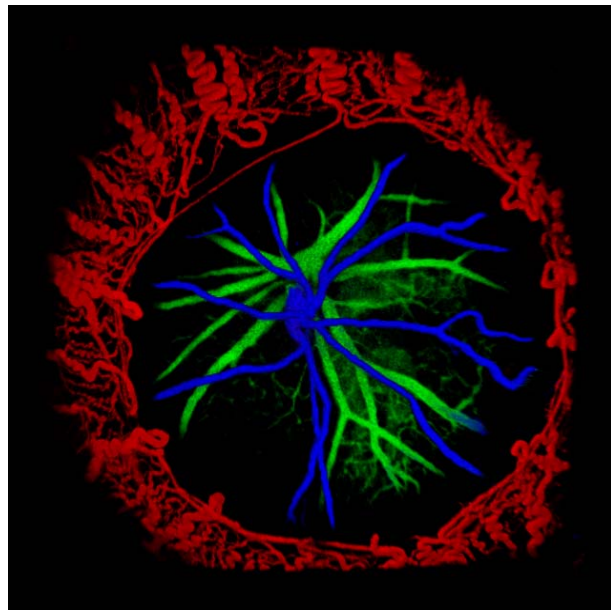


Figure 1: Color depth-coded image of the ocular vasculature rendered in **Volocity Visualization**.

To aid in distinguishing vascular structures, different regions were color coded during image processing according to their distance from the front of the eye .
red=iris vessels; *blue*=hyaloidal vessels;
green=retinal vasculature; scale bar=500 μ m

In order to gain insights into the dynamics of blood vessel development and regression, Professor Martin Friedlander and his colleagues at the Department of Cell Biology have developed a new technique of three-dimensional vascular growth imaging in living animals.

The appropriate guidance and patterning of vessels in the eye during vascular development is critical for proper tissue function. If this patterning, or the vessels themselves, are abnormal, vision can be compromised. In fact, the vast majority of diseases that lead to vision loss do so as a result of abnormal vascular growth. Normal vascular development in the eye is highly regulated and involves both vascular growth and regression. For example, the adult lens is avascular, but during development is supplied by a temporary capillary network, the hyaloidal vasculature and the pupillary membrane (PM). Both vessel networks undergo a natural process of regression over time to ensure the development of a healthy, mature eye with a clear visual axis.

In this study, a minimally invasive method of visualizing the vasculature of the eye *in vivo* was applied. A fluorescently labeled dextran was injected via accessible vessels to visualize the vasculature and changes in the pupillary membrane and hyaloid vessels were observed. Complete regression of the PM was observed at P16 (postnatal day 16), whereas the hyaloid vessels completely disappeared only after 5-6 weeks.

Dr. Ritter and others in the Friedlander laboratory used the advanced rendering techniques of **Volocity Visualization** to create three-dimensional volumes from confocal image stacks as shown in figure 1. **Volocity** was also used to create interactive movie files. **Video 5** at <http://www.iovs.org/cgi/content/full/46/9/3021/DC1> shows the different vascular structures *in vivo*.

The technique of 3D *in vivo* imaging permits the study of vascular development and provides scientists with the possibility of studying angiogenesis under physiologically relevant conditions. In some cases, this method allows the observation of features not possible with other methods because of artifacts associated with dissection or fixation of the tissue. This *in vivo* technique also permits a more physiologically relevant assessment of the effects of drugs on models of neovascular eye diseases and could be a useful surrogate to test pro- and antiangiogenic compounds in general.