AO Wide-Field Microscope

Grant Award Details

AO Wide-Field Microscope

Grant Type: Tools and Technologies I

Grant Number: RT1-01095

Investigator:

Name:	Joel Kubby
Institution:	University of California, Santa Cruz
Type:	PI

Award Value: $549,551

Status: Closed

Progress Reports

Reporting Period: Year 1

View Report

Reporting Period: Year 2

View Report

Grant Application Details

Application Title: AO Wide-Field Microscope

Public Abstract:

A deeper understanding of the biological mechanisms that govern stem cells requires detailed, real-time image analysis of living cells. Currently, conventional live microscopy techniques are ineffective at imaging features like the nucleus in the center of a cell, principally due to aberrations caused by imaging through cytoplasm, organelles and other molecules inside the cell. Similar image distortions occur when looking at an object at the bottom of a pool, where motion of the water causes the image to "shimmer." We therefore propose to develop an improved wide-field microscope with dynamically adjustable optics for live imaging deep within tissues containing stem cells.

Application of technical innovations recently developed by astronomers that provide clearer images of stars is central to our approach. In astronomy, lasers are used to create reference beacons which can be used to adjust the optics in the telescope to correct for image distortions caused by changes in the atmosphere, such as winds and dynamic temperature changes that cause the stars to "twinkle." Such distortions can then be corrected by using a dynamically deformable mirror, similar to the curved mirrors in fun houses, which make you look short or tall. In principle, reference beacons and deformable mirrors can also be used to improve imaging of stem cell samples. The reference beacons control the mirrors that remove warping of the image caused by looking through the materials inside the cell. Our preliminary results indicate small fluorescent beads can be implanted near a feature of interest in cells to serve as beacons to acquire necessary measurements to correct for most distortions.

In year one, we propose to build a device to measure the magnitude of image distortions as a function of depth in living tissue. During this time, we will also design a microscope with sufficient correction to adjust for the measured distortions. In year two, we will build the microscope and characterize its performance. We will test the new imaging system in early fruit fly embryos. With amazing similarity to human cell and developmental biology, these insects are emerging as a key system to safely address many outstanding issues in stem cell biology. Once the imaging system is fine tuned, we will use the technology to examine mechanisms of stem cell self-renewal in the fruit flies.

Once the value of this technology is demonstrated, it will be possible to use it in a wide range of stem cell research, as well as for other applications in cell and developmental biology.

Statement of Benefit to California:	Successful development of an adaptive optics microscope will have an immediate benefit to stem cell research, particularly that research funded by the California Institute for Regenerative Medicine (CIRM). Stem cells usually reside deep within dense cell layers and thus have proven extremely difficult for live fluorescent imaging. Live fluorescent imaging has become an essential tool for revealing the molecular and cell biology of dynamic biological events. With respect to stem cell biology, live cellular imaging will be essential if we are understand how the mechanisms of polarization, asymmetric, self renewal and differentiation. Further, if stem cell biology is to has a therapeutic future this mechanisms must be well understood because we need to control them both in tissue culture as well as in vivo. The adaptive optics scope we are developing should enable us to perform live fluorescent imaging at depths well below the current 30um to 50um limit. This will greatly accelerate research and discovery in fundamental mechanisms of stem cell biology. For example in the studies proposed in this grant, we will be able us to the first time to image live over extended periods, the divisions of germline stem cells of the Drosophila ovary. In addition, we will be able to simultaneously follow the differentiation and of the daughter cells. Once this is achieved this new technology can be readily applied to mammalian stem cell systems. Twenty years ago confocal microscopes were rare. However as they have proven their worthiness and the price has gone down, they are now universal and essential tools in stem cell labs. We envision our adaptive optics microscope as having the same fate. Significantly we are designing the scope so that it can be added on to existing fluorescent scopes thus significantly reducing cost. We hope that within the decade AO microscopes will be a standard tool for stem cell biologists. On a final note, this project will have direct benefit to the California economy as the manufacturing, distribution and selling of the AO scopes would take place in California. We have already had interest from local optics companies in developing the AO scope for market once a suitable prototype is developed.