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| **Prospectus**  Problem to Solve  Increase resolution by reducing point spread function (blurriness)  Image a live sample without inducing photo toxicity.  Significance of Work  Traditional confocal microscopes use a Gaussian beam. A Gaussian beam emits high energy in a focused spot. This can cause biological samples to be destroyed during an experiment.  A Bessel beam is more distributed.  Specimens are imaged by shining a beam on and capturing emission from them.  In this case Caenorhabditis elegans (C. elegans) are imaged to analyze tissue samples, cells, etc.  Methodology   1. Learn LabVIEW 2. Write simple genetic algorithm to optimize **one** image 3. Test on initially blurry image to check if GA is performing as expected (making image sharp) 4. Add 3 image (plane) optimization and work on fitness function 5. System should converge to Bessel beam if optimized **only** on property1 (similarity between planes) 6. Combine different sets of properties    1. Similarity between planes    2. Size of laser beam    3. Maximum of peaks 7. Create line focus | **Proposal**  Problem to Solve  Increase resolution by reducing point spread function (blurriness)  Image a live sample without inducing photo toxicity.  Background  **Previous work** – Bessel beams are used in conjunction with structured illumination and/or two-photon excitation to create thinner light sheets better suited to three-dimensional subcellular imaging. Currently, microscope is capable of noninvasively acquiring hundreds of 3D isotropic data volumes from single living cells encompassing tens of thousands of image frames.  **Significance of Work** –  Feasibility  Show that Bessel beam imaging is a viable method for noninvasively acquiring data from a live specimen.  Timeline   1. Learn LabVIEW 2. Write simple genetic algorithm to optimize **one** image 3. Test on initially blurry image to check if GA is performing as expected (making image sharp) 4. Add 3 image (plane) optimization and work on fitness function 5. System should converge to Bessel beam if optimized **only** on property1 (similarity between planes) 6. Combine different sets of properties    1. Similarity between planes    2. Size of laser beam    3. Maximum of peaks 7. Create line focus   Budget  **Hardware** –  **Software –** MatLab, LabVIEW, Windows 7  References |
| **Article Extracts**  **Abstract** – *Rapid three-dimensional isotropic imaging of living cells using Bessel beam plane illumination*  Thomas Planchon, Liang Gao, Daniel E Milkie, Michael W Davidson, James Galbraith, Catherine Galbraithb, Eric Betzig  A key challenge when imaging living cells is how to noninvasively extract the most spatiotemporal information possible. Unlike popular wide-field and confocal methods, plane-illumination microscopy limits excitation to the information-rich vicinity of the focal plane, providing effective optical sectioning and high speed while minimizing out-of-focus background and premature photo bleaching. Here we used scanned Bessel beams in conjunction with structured illumination and/or two-photon excitation to create thinner light sheets better suited to three-dimensional subcellular imaging. As demonstrated by imaging the dynamics of mitochondria, filopodia, membrane ruffles, intracellular vesicles and mitotic chromosomes in live cells, the microscope currently offers 3D isotropic resolution, speeds up to nearly 200 image planes per second, and the ability to noninvasive acquire hundreds of 3D data volumes from single living cells encompassing tens of thousands of image frames.  Adaptive Optics Is a process used to enhance the performance of an optical system by reducing its **wavefront distortion**. It is used ext4ensively in astronomy, ibn addition to numerous other applications within the field of optics; such as optometry and microscopy.  Bessel Beams have minimal diffraction, and when focused, allow for a long and narrow beam that is essential for **Light-Sheep Microscopy**. The output spatial profile is typically that of a Gaussian beam. Hence, to obtain a Bessel Beam, the initial spatial profile needs to be transformed/shaped. This project aims to explore the possibility of engineering a Bessel beam profile through the use of a bi-dimensional Spatial Light Modulator.  Due to the statistical nature of the photons generated in the laser cavity, the output laser has a spatial profile that is very close to a Gaussian beam. The goal of the experiment is to transform this Gaussian profile into a Bessel beam profile. | **Jargon**  **Diffraction** - refers to various phenomena which occur when a wave encounters an obstacle or a slit. In classical physics is described as the interference of waves according to the Huygens–Fresnel principle.  **Diffraction limit** – lowest resolution you can get  [**Biophotonics**](http://photonicssociety.org/newsletters/apr04/biophoto.html) – Use of light (photons) to image, detect and manipulate biological materials.  [**Phototoxicity**](https://en.wikipedia.org/wiki/Phototoxicity) – chemically induced epidermal irritation requiring light  **Point spread function** – how well you can see two nearby objects before they become one object |

**Break Down of Genetic Algorithm**

**Population** – potential solutions (parameters)

**Simulation** - inputs parameters and gives outputs (models)

**Fitness Function** - Evaluates the performance of a solution

After an evaluation, the most *fit* solution “survives”. The least *fit*, “dies” off.

The most fit functions are fed back into the population and mutated (bred, or hybridized).

(Their parameter values are mixed and matched)

The new entity in the population is fed back into the simulation and the process is repeated

**Other Notes**

**GA Objective -** Calibrate microscope to optimize focal length.

**Bessel beam** is low intensity radiation (relative to Gaussian)